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نموذج رقم (١٨)
اقرار والتزام بقوانين الجامعة الأردنية وأنظمتها وتعليماتها لطلبة الماجستير

أنا الطالبة: مروة نعيم خليل أبو عيشة الرقم الجامعي: 8081531

التخصص: الكيمياء الكلية: العلوم

اعلن بأنني قد التزمت بقوانين الجامعة الأردنية وأنظمتها وتعليماتها وقراراتها السارية المفعول المتعلقة باعداد رسائل الماجستير والدكتوراة عندما قمت شخصيا" باعداد رسالتي / اطروحتي بعنوان: Synthesis and Bioassay of some N-(Flavon-7-yl) amidrazones and related congeners.

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توقيع الطالب: .... التاريخ: / /

SYNTHESIS AND BIOASSAY OF SOME N1-(FLAVON-7-YL) AMIDRAZONES AND RELATED CONGENERS

By
Marwa Naim Abu-Aisheh

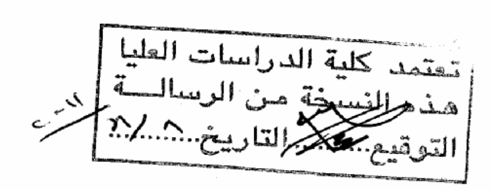
Supervisor
Dr. Mohammad S. Mubarak, Prof.

Co-supervisor
Dr. Mustafa M. El-Abadelah, Prof.

This Thesis was submitted in Partial Fulfillment of the Requirements For the Master's Degree in Chemistry

Faculty of Graduate Studies
The University of Jordan

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Committee Decision

This Thesis (Synthesis and Bioassay of some N1-(flavon-7-yl) amidrazones and related congeners.) was successfully defended and approved on Tuesday 12-07-2011

Examination Committee
Signature

Dr. Mohammad S. Mubarak, Chairman
Prof. of Organic Chemistry

Dr. Mustafa M. El-Abadelah, Co-Chairman
Prof. of Organic Chemistry

Dr. Musa H. Abu Zarga, Member
Prof. of Organic Chemistry

Dr. Haythem A. Saadeh, Member
Associate. Prof. of Organic Chemistry

Dr. Mahmoud Y. Al-Talib, Member
Prof. of Organic Chemistry
(Yarmouk University)

Dedication

To all my beloved

To My father, mother, brother

Sisters, friends

Whom I owe a lot,

I dedicate this thesis

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All Praise towards Almighty Allah who has been my source of strength and to whom I owe all that I have been able to do and accomplish.

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2D : Two Dimensional.

COSY : Correlated Spectroscopy.

DEPT : Distortionless Enhancement by Polarization Transfer.

HIV : Human Immunodeficiency Virus.

HMBC : Heteronuclear Multiple Bond Connectivity.

HMQC : Heteronuclear Multiple Quantum Coherence.

HRMS : High Resolution Mass Spectrometry.

MS : Mass Spectrometry.

NMR : Nuclear Magnetic Resonance.

TLC : Thin Layer Chromatography.

IC_{50}: Half maximal inhibition concentration.

Synthesis and Bioassay of some N1-(flavon-7-yl) amidrazones and related congeners

By

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Abstract

The new flavone-7-yl hydrazonoyl chloride (**78**) was obtained by the Japp-Klingemann reaction, starting from 7-amino-4-flavone. This new hydrazonoyl chloride was reacted with selected secondary amines in basic media, to deliver the corresponding flavone-7-yl amidrazones (**79a-n**). New flavone-7'-yl [1,2,4]triazin-6-ones compounds (**80a-k**) and flavone-7'-yl pyrrolo[1,2-*d*][1,2,4]triazin-1-one **(81)** were synthesized by treating flavone-7-yl hydrazonoyl chloride with L-(α)-amino acid methyl esters. These newly synthesized compounds were characterized by different spectroscopic techniques, such as ^{1}H-NMR,^{13}C-NMR, DEPT, 2D-NMR (COSY, HMQC and HMBC) and mass spectrometry. Preliminary screening of these compounds Against different tumer cell lines was performed ; and a number of these compounds exhibited good to significant antitumor activity against T47D, MCF-7 breast cancer and K562 leukemia cell lines.

1. Introduction

1.1. Importance and applications of Chromones

Chromone **1**, 4H-chromen-4-one, is a 4H-1-benzopyran-4-one (Ellis, 1977), (Sosnovskikh, 2003), (Buchholz *et al*., 2005), that is a derivative of benzopyran **2** with a substituted keto group on the pyran ring (Altounyan and Howell, 1967).

Chromone crystallizes in colorless needles of mp 59 °C (Becker, 1991), (Eicher and Hauptmann, 2003). The first chromone used in pure form in clinical practice was khellin **3**, which was extracted from the seeds of the plant *Ammi visnaga* (Mustapha, 1879), (Edwards and Howell, 2000).

Chromones have attracted much synthetic interest because of their reactivity and their biological activity of its naturally-occurring representatives (Bracke, *et al.,* 1997), (Marder, *et al.,* 1998),(Bhat, *et al.,* 1999), (Miao and Yang, 2000), (Mubarak and Ayoub,

2007).Chromones are well known for their antioxidant (Jovanovic, *et al*., 1994) (Kumar and Yusuf, 2006), anti-inflammatory, antifungal, antimicrobial, antiviral, antitumor and anticancer activity due to their well-recognized antioxidant properties, stem from their ability to neutralize active forms of oxygen and to cut off free radical processes. (Machado, 2010).

1.2. Natural chromones

Molecules containing the chromone ring have been the subject of considerable chemical interest in the past decades. They occur widely in nature and exhibit important biological as well as pharmacological activities (Billet, *et al*., 1945), (Azima, *et al*., 1951), (Cox, 1967), (Cox, *et at*.,1970), (Kumar and Yusuf, 2006). They have been shown to be tyrosine and protein kinas C inhibitors, as well as antifungal, antiviral, antitubulin, antihypertensive (Bhat, *et al.,* 1999) and anticancer agents (Bracke, *et al.,* 1997), These compounds also possess low mammalian toxicity and are present in large amounts in the diet of humans due to their origin in plants (Hoult, *et al*., 1994), (Bourne *et al*., 2003).

Recently, some chromones are reported as anti-HIV agents (Alloway, *et al*., 2003); Khellin **3** **(**Brehm, et al., 1994) and 2, 4-thiazolidenedione **4** **(**Dow and Kreutter,1995), (Bhushan, *et al*., 1998), (Ishar, *et al*., 2002) were used as antispasmodic agents in the treatment of anginapectoris and antidiabetic agents (Kumar and Yusuf, 2006).

4

Y = S, NH, NMe, NEt ; Z = O, S

New Chromone glucosides, takanechromones (**5 – 7**), were isolated from the methanolic extracts of *Hypericum sikokumontanum* together with 27 known compounds. The isolated compounds and some chromone derivatives were assayed for antimicrobial activity against *Helicobacter pylori* and cytotoxicity against human cancer cell lines. (Higuchi *et al*., 2009)

	R
5	**Me**
6	**Et**

7

1.3. Synthesis of chromones

Most syntheses of chromones require the prior construction of a 1-(ortho-hydroxyl)-1,3-diketone **9**, and it is in the manner in which this intermediate is generated that the methods differ (Joul and Mills, 2000). Such methods include: Claisen condensation, Baker-Venkataraman rearrangement (Eicher and Hauptmann, 2003), and Kostanecki-Robinson reaction (Wawzonek, 1950).

1.3.1. Claisen condensation

The most frequently used method is the acid-catalysed cyclization of o-hydroxyaryl-1, 3- diketones **9**, which are obtained from o-hydroxyacetophenones **8**, especially in their O-silyl (Cushman and Nagarathan, 1991) protected form, by a Claisen condensation, (Eicher and Hauptmann, 2003).

1.3.2. Baker-Venkataraman rearrangement

An alternative route to ß-diketones **9** is the base-catalysed isomerization of o-acyloxy acetophenones **11** *,* which are readily obtained by O-acylation of o-hydroxyaceto phenones:

The Baker-Venkataraman rearrangement can be regarded as a 1, 5-acyl migration in the enolate **12** and is of great value in the synthesis of flavones (Eicher and Laas, 1989), (Eicher and Hauptmann, 2003).

1.3.3. Kostanecki-Robinson reaction

This reaction is a reliable method for the preparation of chromone only from aromatic anhydrides and the corresponding sodium salts. This combination gives flavones and has probably been used more frequently for the preparation of these compounds than any other method because of the simple steps involved. Cinnamic acid **13** derivatives behave similary and give the 2-styrylchromones **14**.(Wawzonek, 1950).

HO OH R O 13

$(C_6H_5CH = CHCO)_2O$

$C_6H_6CH = CHCOONa$

180 - 190 °

R' O O O R O 14

1.4. Reactions of chromones

Chromones show analogies in their reactions to 4H-pyran-4-one, i.e. they behave as masked 1, 3-dicarbonyl systems (Eicher and Hauptmann, 2003). Protonation, alkylation, reaction with electrophilic and nucleophilic reagents, reaction with oxidizing agents (Joul and Mills, 2000) and photo-structural transformations. (Kumar and Yusuf, 2006).

1.4.1. Protonation and alkylation

Protonation and alkylation occur on the carbonyl oxygen to produce a hydroxylbenzo-pyrylium salt **15**. (Joul and Mills, 2000)

O O

Et_2O

HCl

OH $\overset{+}{O}$ Cl^-

15

1.4.2. Reaction with electrophilic reagents

Electrophilic attack takes place at the deactivated pyran-4-one ring in the 3-position; e.g. aminomethylation **16** can be brought about under Mannich conditions. (Eicher and Hauptmann, 2003).

O
O
CH_2O, Me_2NH, HCl
EtOH, reflux, 60%
O
CH_2NMe_2
O
16

1.4.3. Reaction with nucleophilic reagents

The chromone system behaves as a Michael acceptor towards nucleophiles. Normally, attack occurs at C-2, but is less likely on C-4. The addition product underwent to frequently ring transformations. (Eicher and Hauptmann, 2003). Ring-opened products **17** was obtained from the reaction of chromones and secondary amines where the nucleophilic agent has attacked at C-2. (Joul and Mills, 2000)

O
O
Et_2NH
EtOH, RT
O^-
H
O
NEt_2
O
NEt_2
H
OH
17

1.4.4. Reaction with oxidizing agents

2-arylchromone (Flavones), are quantitatively convertd into 2,3-epoxide **18** by exposure to dimethyl dioxarine. This synthetic intermediate is converted by acid to 3-hydroxy flavones **19**, which are naturally occurring (Adam, et al., 1991), (Adam, et al., 1992), (Joul and Mills, 2000)

1.4.5. Photo-structural transformations of chromone

Chromones are bichromophoric substrates that contain double bond as well as C=O group as the chromophoric units which can undergo photo-excitation either in isolation or in conjugation. Chromones undergo photocycloaddition, photodimerisation, photoisomerisation, photorearararrangement, photooxidation-reduction and photocyclisation reactions involving both n→π* and π→π* transitions. For an example: Gupta and Mukerjee (Gupta and Mukerjee, 1973) have reported the phototransformation of 3-methoxychromone **20** where H-abstraction coupled with dimerization has led to the formation of dimeric oxetanol **21**.(Kumar and Yusuf, 2006).

1.5. Importance and application of Flavonoides

Chromone, the 4H-[1]-benzo-4-pyranone system; is the basis of the chemical structure of a large group of biologically active natural products known as flavonoids and isoflavonoids. (Frasinyuk and Khilya, 1999)

Flavonoids, a group of benzopyrone derivatives, (Bonina *et al.*, 1995) have been recognized as one of the largest and most widespread class of plant constituents occurring throughout the plant kingdom (Ducrot, 2007). Flavonoides such as flavones are present in a great varity of foods, and specially in fruits and vegetables (Artusi, 2009). Flavonoids have been shown to possess several biological properties including hepatoprotective, anti-thrambotic, antiinflammatory, and antiviral activities. Many of which may be related, partially at least, to their antioxidant and free-radical-scavenging ability (Gryglewski and Robak 1988), (Chen et al., 1990), (Bonina *et al.*, 1995)

1.5.1 Structure of flavonoides

The basic structural feature of flavonoid compounds is the 2-phenyl-4H-1-benzopyran-4-one nucleus, consists of two benzene rings (A and B) linked through a heterocyclic pyran ring (C), **22** (Brown, 1980) (Cushnie and Lamb, 2005).

O 4 3 5 4a 6 A C 2 2' 7 8a 1' 3' 8 O B 6' 4' 22 5'

The individual carbon atoms are based on a numbering system which uses ordinary numerals for the A and C and "primed" numerals for B-ring **22** (Havsteen, 1983). The different ways to close this ring associated with the different oxidation degrees of ring A provide the various classes of flavonoids. The six-membered ring condensed with the benzene ring is either a pyrone (flavones **22,** flavonols **23**) or its dihydroderivative flavanones **24** and flavan-3-ols **25** (Baxter and Harborne, 1999), (Sakarkar *et al.*, 2008).

O OH O 23

O O 24

OH O 25

1.5.2. Importanc and applications of Flavones

Chromones with a 2- phenyl susbstituent have the trivial name flavones **22**. (Gilchrist, 1997). The 2-phenyl-4H-1-benzopyran-4-one nucleus is well known in naturally occurring compound, as class of benzopyran-4-ne derivatives. Flavones belong to a very important class of natural compounds of the flavonoid group and are widely occured in nature. Flavones are present in a great varity of food, especially in fruits and vegetable (Artusi *et al*., 2009).

A great number of biological activities were reported for flavones (Kasum and Ross, 2002), (Blumberg *et al.*, 2005), such as anti-estrogenic (Belluti *et al*., 2006) , anti- tumor (Azqueta et al., 2007), anti-allergic (Arimitsu *et al*., 2007), or anti-inflammatory (Biswas *et al*., 2006), (Crespo et al., 2007). These promising properties led to numerous chemical works focusing on the synthesis and the structural modifications of flavones (Carballido-Reboredo *et al*., 2005), (Kabalka and Mereddy, 2005), (Dahlen *et al*., 2006), (Dubrovsky and Larock, 2006), (Dixon *et al*., 2007), (Kumar and Perumal, 2007), (Demuynck, 2008).

1.5.3. Natural flavones

Most naturally occurring flavones are hydroxylated at positions 5 and 7. An example is luteolin **26**, a yellow compound which was obtained from *Reseda luteola* (wild wood) and used as dyestuff. A related pentahydroxyflavone is quercetin **27**, which is one of the most widely, distributed natural yellow pigments. It often occures in the form of glycosides. (Acheson, 1976)

1.5.4. Synthesis of Flavones

Flavone can be synthesized by a number of methods:

1.5.4.1. Claisen condensation

Intramolecular Claisen condensation between an ester group and the methylene adjacent to the carbonyl of the acrylarene **28** produces a 1-(ortho-hydroxyaryl)-1,3-diketone **29**. The claisen condensation can be conducted in the presence of the acidic phenolic hydroxyl by the use of excess base. Alternatively the process is conduted in two steps: first acylation of the phenolic hydroxyl, secondly, an intramolecular (Baker, 1933), (Banholzer, and Schmid, 1954) base-catalysed Claisen condensation, known as Baker-Venkataraman rearrangement. (Joul and Mills, 2000)

1.5.4.2. Kostanecki-Robinson reaction

Preparation of flavones **31** from *o*-hydroxyaryl ketones **30** with anhydrides of aromatic acids and their sodium salts (Allan and Robinson, 1924).

1.5.4.3. From chalcones

A versatile flavone synthesis consists of the oxidative cyclization of chalcones **32** by selenium dioxide in higher alcohols:

It involves an intramolecular, possibly acid-catalysed, Michael addition of the phenolic OH group of 32 followed by a cyclization to the flavanone **33,** which undergoes dehydrogenation to give a flavones **22**. (Eicher and Hauptmann, 2003).

1.5.5. Reactions of Flavones

The chemical behavior of flavones, and their derivatives is due to their polyfunctional nature. Flavone compounds can enter into the most diverse reactions: electrophilic substitution, oxidation, reduction, cycloaddition, condensation, recyclization, and many others. (Ishchenko and Khilya, 2002). Flavones show analogies in their reactions to chromones (Eicher and Hauptmann, 2003) (see 1.4).

1.5.6. Biological activity of Flavonoids

Flavonoids exhibit wide range of biological activities arising mainly from their antioxidant properties and ability to modulate several enzymes or cell receptors.(Hodek *et al*., 2002) They have been reported to possess many useful properties, including anti-inflammatory activity, enzyme inhibition, antimicrobial activity (Havsteen, 1983), (Baxter and Harborne, 1999), antiallergic activity, antioxidant activity (Chithan and Middleton, 1993), anti-HIV, vascular activity and cytotoxic antitumour activity (Harborne and Williams, 2000) . (Cushnie and Lamb, 2005)

1.5.6.1. Antioxidative effects of flavonoids

The best-described property of almost every group of flavonoids is their capacity to act as antioxidants. The flavones and catechins seem to be the most powerful flavonoids for protecting the body against reactive oxygen species. Body cells and tissues are continuously threatened by the damage caused by free radicals and reactive oxygen species, which are produced during normal oxygen metabolism or are induced by

exogenous damage (de Groot, 1994), (Grace, 1994). Flavonoids can prevent injury caused by free radicals and reactive oxygen species, by scavenging of free radicals. Flavonoids are oxidized by radicals, resulting in a more stable, less-reactive radical. In other words, flavonoids stabilize the reactive oxygen species by reacting with the reactive compound of the radical (Boelens et al., 2001).

1.5.6.2. Antifungal activity of flavonoids

Owing to the widespread ability of flavonoids to inhibit spore germination of plant pathogens, they have been proposed for use against fungal pathogens of man (Harborne and Williams, 2000). Two new flavones from *Artemisia giraldi*, identified as 6,7,4-trihydroxy-3',5'-dimethoxy flavones **34** and 5,5'dihydroxy-8,2',4'-trimethoxyflavone **35**, together with 5,7,4'- trihydroxy-3',5'-dimethoxyflavone **36** have been reported to exhibit activity against *Aspergillus flavus* (Tan *et al*., 1996), a species of fungi that causes invasive disease in immunosuppressed patients (Harley and Klein, 1999), (Cushnie and Lamb, 2005).

1.5.6.3. Anti-inflammatory effects of flavonoids

Flavonoids inhibit cyclooxygenase and lipoxygenase which play an important role as inflammatory mediators. Quercetin **27**, in particular, inhibits both cyclooxygenase and lipoxygenase activities, thus diminishing the formation of these inflammatory metabolites (Gryglewski and Robak, 1996), (Iversen *et al*., 1998), (Boelens *et al*., 2001).

1.5.6.4. Antiviral effects of flavonoids

Flavonoids also have inhibitory activity against a variety of viruses. For example, Selway reports that quercetin **27**, morin **37**, dihydroquercetin **38**, dihydrofisetin **39**, leucocyanidin **40**, pelargonidin chloride **41** and catechin **42** possess activity against up to

seven types of viruses, including herpes simplex virus (HSV), respiratory syncytial virus, poliovirus and Sindbis virus (Selway, 1986), (Chithan and Middleton, 1993). In addition, it has been demonstrated that several flavonoids, including demethylated gardenin A **43** and 3, 2-dihydroxyflavone **44**, Robinetin **45** inhibit HIV-1 proteinase (Brinkworth, 1992), (Cushnie and Lamb, 2005).

1.5.6.5. Anti-bacterial activity of flavonoids

Several flavonoids exhibit anti-bacterial activities. Experiments with bacteria showed that myricetin **46** inhibits the growth of multidrug resistant *Burkholderia cepacia*, vancomycin-resistant enterococci and other medically important microorganisms, such as *Klebsiella pneumoniae* and *Staphylococcus epidermidis* (Lee, and Xu, 2001), (Hodeke *et al*., 2002).

1.5.6.6.Cytotoxic antitumor activity of flavonoids

Among flavonoids biological properties, antitumor activities and antiproliferative effects have aroused considerable attention (Chabot *et al*., 1997). The essential feature of flavonoids is their free radical scavenging activity. These antioxidant properties are, in part, responsible for their antitumor effects. They prevent cell damage caused by reactive oxygen species formed via normal metabolic processec(Capasso *et al*., 1999), (Daniel *et al*, 1999), (Boelens *et al*., 2001). Flavonoid compounds were repoterd to be cytotoxic for cancer cells but not for normal cells. (Kosmider and Osiecka, 2004).

1.5.6.7. Antitumor activity of Flavones

Flavone (2-phenyl-4H-1-benzopyran-4-one), the non-hydroxylated core structure of the flavones subgroup, have several biological activities, incuding antitumor activity which is one of the promising studies (Beney *et al*., 2003). Flavone proved to be a stronger apoptosis inducer than the clinically established antitumor agent camptothecin, a topoisomerase I inhibitor which is usually applied as a second-line pharmacotherapeutic in advanced colorectal cancers promote apoptosis (Brendel *et al*., 2000), (Martens, 2005), for an example: flavone-8- acetic acid **47** posses antitumor properties (Cradock *et al*., 1986), (Eicher and Hauptmann, 2003).

1.5.7. Amino flavones

Substituted flavones are highly attractive derivatives due to their therapeutic potential. The substitution pattern of these compounds is crucial for their biological activity. Literature survey regarding the structure-activity relationship of flavone, indicates that: azaflavones are highly active molecules, positions 5 and 7 are the most important; hydroxyls, methoxy and amino groups are the most beneficial (Beney, 2003). Flavonoids; bearing amino groups on the **A** or **B** ring have been reported to be potential antineoplastic agents (Akinaga *et al*., 1990), (Gomi *et al*., 1990), (Cushman *et al*., 1991), (Cunningham *et al*., 1992), (Akama *et al*., 1993), (Cushman *et al*., 1994). It is now well established that such potency is mainly due to the ability of these aminoflavones to be competitive inhibitors of certain protein tyrosine kinases with respect to ATP (Cuhman, 1991), (Cunningham *et al*., 1992), (Cwhman *et al*., 1994), (Chabot *et al*., 1997)

1.6. Amidrazones

Amidrazones are weak monoacid bases characterized by the structural formula **48**, where R, R', R'', R''' and R'''' can be any of a wide variety of atomic or organic moieties. A particularly well known example of this class of compounds is aminoguanidine **49**. (Aly and Nour-El-Din, 2008)

In the free state, amidrazones tend to be either liquids or low-melting solids, (Aly and Nour-El-Din, 2008). Amidrazones are able to exhibit tautomerism, some amidrazones exist in an amide hydrazone structure while others are exclusively in hydrazide imide form.(Bahceci et al., 1999)

1.6.1 Importance of amidrazones

Amidrazones are known as convenient building blocks for various N-heterocycles, such as 1,2,4-triazoles (**50**) or 1,2,4-triazines (**51**) (Katritzky *et al.*, 1979).

In recent years, various biological activities have been discovered for amidrazone compounds, e.g., fungistatic , bacteriostatic, and antimycotic activities as well as inhibitory effects on mammalian and plant enzymes, e.g. lipoxygenases $(LOX)_4$ that are possibly subject to a redox mechanism.

Several compounds containing an amidrazone moiety are known to be potent inhibitors of lipoxygenase-1 activity. Recently, (Clemens, *et al*., 2001) reported that compounds **52** and **53** acts as lipoxygenase-1 inhibitors with the half maximal inhibitory concentration (IC_{50})-values of 10 and 38 nM, respectively.

52 **53**

On the other hand, new piperazinyl amidrazones were synthesized *via* direct interaction of the corresponding arylhydrazones with the appropriate piperazine e.g. compounds **54 a, b** (Abdel-Jalil *et al*., 2010). Piperazine-based compounds have been employed as antibacterial (Khalaj *et al*., 2004), antidepressant (Broekkamp *et al*., 1995), antitumor drugs (Naito *et al*., 2005), as d-adrenoceptor antagonists (Ibarra *et al*., 2000), CCR_5 receptor antagonists (Jiang *et al*., 2004), 5-HT7 receptor antagonists (Yoon *et al*., 2005), and adenosine A_2A receptor antagonists (Vu *et al*., 2004).

54 a: R= Me
54 b: R= Et

1.6.2. Nitrile imines

Nilrile imines are 1,3-dipolar species (Heusgen, 1968), which could be represented by the following resonance structures:

Nitrile imines are thermally unstable; therefore they could be generated *in situ* from their precursors in the presence of a suitable reactant (Shawali and Parkanyi, 1980). Nitrile imines are considered as important starting intermediates for the synthesis of varios amidrazones. Nitrile imines could be prepared by various methods such as: Elimination of hydrogen halide from hydrazonoyl halides **55** by a base (Butler and Scott, 1970).

RC=N—NAr (X, H) **55** → Base, -HX → $RC\equiv\overset{+}{N}—\overset{-}{N}Ar$ **56**

Hydrazonoyl halide, are considered important compounds to generate the respecive nitrile imine; which could be prepared by several routes such as: Japp-Klingimann reaction.

1.6.3. Japp-Klingimann reaction

This reaction is considered to be the most important method for the synthesis of hydrazonoyl halides **58**. It is accomplished by coupling of aryl diazonium salts **57** with methinyl compounds which are activated by two electron-withdrawing groups in basic aqueous media, such as sodium acetate or pyridine, giving high yields of hydrazonoyl halides (Phillips, 1959).

R_1 $\overset{+}{N}_2Cl$ + R_2 HC—X R_3 Base /H_2O → [R_1 N=N—C—X (R_2, R_3)] → R_1 $\overset{H}{N}$—N=C (X, $R_2(R_3)$)

57 **58**

R_2,R_3:(Ac, CO_2Et, CN, *etc.*)

1.6.4. Reactions of nitrile imines

Nitrilimines react with two modes which are:

1.6.4.1. 1,3-dipolar cycloaddition reactions

Nitrile imines **59** react with various dipolarophiles **60** such as alkene, alkyne, aldehydes, ketones, thioketones and nitriles (Shawali, 1993).

1.6.4.2. Nucleophilic addition reactions

Nitrile imines (**59**) are susceptible to attack by different nucleophilic species such as water, alcohols (Butler and Scott, 1970), amines (Sharp and Hamilton, 1946), thiolates (Wolkoff, *et al*., 1974), etc.

In particular, primary and secondary amines add readily to nitrile imines, the interaction between nitrile imines and the amino component is expected to yield the corresponding Z-amidrazones **65** as the kinetically controlled products (Hussein *et al*.,1984).

When the reacting nucleophile contains a suitably located electrophilic center, intracyclization, following the addition step, can occur (*via* the nitrile imine nitrogen and the electrophilic center), leading to a five-membered ring (Shawali and Abdelhamid, 1976) or six-membered ring.

The reaction of α-amino esters with nitrile imines was described (El-Abadelah *et al*. 1991), in which α-amino esters add through the amino group onto nitrile imines (in a similar manner to amines) to give the corresponding Z-amidrazone esters **66**, which can undergo a spontaneous cyclocondensation reaction to afford (by elimination of alcohol) 1,2,4-triazin-6-one (**67**). This type of intracyclization is classified as an allowed "6-Exo-Trig" process (Baldwin, 1976).

1.6.5. Triazines (4,5-dihydro-1,2,4-triazin-6-ones)

Triazines **68-70** are 6-membered heterocycles-triazabenzenes. The chemistry, and utility of 1,2,4-triazines **70** have been extensively studied and reviewed (Raw and Taylor, 2010).

1,2,3-trazine 68 | **1,3,5-trazine 69** | **1,2,4-triazine 70**

4, 5-Dihydro-1, 2, 4-triazin-6-one **71** is a derivative of 1,2,4-Triazin-6-one **72**; both of which are considered as a cyclic amidrazones.

1.6.5.1. Synthesis of 4, 5-dihydro-1, 2, 4-triazin-6(1*H*)-one

The first synthesis of 4,5-dihydro-1,2,4-triazin-6-one was reported from reaction of 1-(N-phenylglycyl) phenyl hydrazine (**73**) with formic acid giving 1,4-diphenyl-1,2,4-triazin-6-one (**74**) (Widman, 1893).

Later, 5-substituted 4,5-dihydro-1,2,4-triazin-6-ones became available by different routes such as: Reaction of hydrazonoyl chloride (**75**) with α-amino acid esters afford 4,5-dihydro-1,2,4-triazin-6-ones **76** (El-Abadelah *et al*., 1991).

2. Purpose of the present work

Based on the properties of flavones and amidrazones (cited in the introduction), we plan to synthesize a set of new N1-(flavones-7-yl) amidrazones and N1(flavone-7-yl)4, 5-dihydro [1,2,4]triazin-6-ones in search for possible anticancer agents.

Work strategy

1. Synthesis and characterization of 2-oxo-N-(4-oxo-2-phenyl-4*H*-chromen-7-yl) propanehydrazonylchloride (**78**).

i) $NaNO_2$ / 6 N HCl
ii)$(Ac)_2CHCl$ / aq. NaOAc / EtOH

77 → **78**

2. Synthesis and characterization of new flavone-7-yl amidrazones (**79a-n)**.

NEt_3 / DMF

78 → **79a-n**

Compounds **79a-n**

entry	a	b	c	d	e	f	g	h	i	j	k	l	m	n
X	CH_2	S	O	NH_2	NMe	NEt	NBz	N(2-pyrimidine)	N(p-Ph)	N(p-C_6H_4OMe)	N(o-C_6H_4F)	N(p-C_6H_4F)	N(CO_2Et)	N(p-C_6H_4Cl)

3. Synthesis and characterization of new flavone-7'-yl 4,5-dihydro-1,2,4-triazin-6-ones (**80a-k**)

Ac Cl N N H 78 O O Ph + R_2HN O O Me R_1 NEt_3 / EtOH -5 to 0 C Ac N N N R_2 R_1 O O O Ph 80a-k

Compounds **80a-k**

	R1	R2
a	$CH_2CH(CH_3)_2$	H
b	CH_3	H
c	H	CH_3
d	H N (indoline) $^{-}CH_2$	H
e	$CH_2CH_2SCH_3$	H
f	CH_2Ph	H
g	$CH_2CH_2CO_2Me$	H
h	H	H
i	CH_2OH	H
j	$CH(OH)CH_3$	H
k	CH_2CO_2Me	H

4. Preparation of and characterization of flavone-7'-yl pyrrolo[1,2-d][1,2,4]triazin-1-one **(81).**

Ac, Cl, N, N, H, O, O, Ph, **78** + R_2HN, O, O, Me, R_1 — NEt_3 / EtOH, -5 to 0 C → Ac, N, N, N, R_2, R_1, O, O, O, Ph, **80a-k**

5. Evaluate the anticancer activity for newly synthesized compounds.

3. Experimental

3.1. Materials and equipments

All chemicals used were obtained from commercial sources and were used as received without further purification. Piperidine, morpholine, thiomorpholine, piperazine, N-alkylpiperazines, N-arylpiperazines, N-acylpipeprazines, pyrrolidine were purchased from Acros. (L)-α-Amino acids methyl esters of (alanine, leucine, phenylalnine, glycine, tryptophane, proline, methionine, sarcosine, serine and therionine), (L)-α-amnio acid dimethyl esters of aspartic and glutamic acids were obtained from Aldrich. Silica gel for column chromatography was recieved from Macherey-Nagel GmbH & Co (Germany). Melting points (uncorrected) were determined on a Stuart scientific melting point apparatus in open capillary tubes. Optical roations were taken on a Perkin Elmer 141 photoelectric spectropolarimeter in dimethylformamide ($c \sim 1$), at 20 ± 1 ^{o}C. ^{1}H- and ^{13}C-NMR spectra were recorded on a 300 MHz spectrometer (Bruker DPX-300) with *TMS* as the internal standard. Chemical shifts are expressed in δ units; ^{1}H-^{1}H, ^{1}H-F and ^{13}C-F coupling constants are given in Hertz. High resolution mass spectra (HRMS) were acquiered by electrospray ionization (ESI) technique with the aid of Bruker APEX-2 instrument. The samples were dissolved in acetonitrile, diluted in spray solution (methanol / water 1:1 v/v + 0.1 % formic acid) and infused using a syringe pump with a flow rate of 2 μL / min. External calibration was conducted using arginine cluster in a mass range *m/z* 175-871.

3.2. Synthesis

3.2.1. Preparation of 2-oxo-N-(4-oxo-2-phenyl-4*H*-chromen-7-yl) propanehydrazonylchloride (78).

The title compound was prepared by following two procedures:

Procedure I

Step (i). 7-Amino-flavone **77** (23.7g, 0.10mol) was dissolved in 17% aqueous hydrochloric acid (160 mL). To this solution was added, dropwise, a solution of sodium nitrite (8.0 g, 0.11 mol) in water (15 mL) with efficient stirring at 0-5 °C. Stirring was continued for 20-30 min., and the resulting fresh cold 4-oxo-2-phenyl-4*H*-chromen-7-diazonium chloride, [7-(chlorodiazenyl)-flavone], solution was used immediately for the following coupling reaction.

Step (ii). A cold (-5° C) freshly prepared solution of 4-oxo-2-phenyl-4*H*-chromen-7-diazonium chloride (0.1 mol) was poured onto cold solution (0 to -10 °C, ice-salt bath) of 3-chloropentan-2,4-dione (13.4 g, 0.1 mol) in pyridine / water (160 ml , 3:2 v/v) with vigorous stirring. The resulting yellow-colored mixture was further stirred until a solid precipitate was formed (5-10 min). The reaction mixture was then diluted with cold water (200 mL), the solid product was collected by suction filtration, washed several times with cold water, dried, and recrystallized from ethanol . Yield 33.0 g (97.2 %).

Procedure II

Step (i).the same as the step (i) mentioned above.

Step (ii). A cold (-5°C) freshly prepared solution of 4-oxo-2-phenyl-4H-chromen-7-diazonium chloride (0.1 mol) was poured onto cold solution (0 to -10 °C, ice-salt bath) of 3-chloropentan-2, 4-dione (13.4 g, 0.1 mol) in Ethanol / water (160 ml, 1:1 v/v) containing 20.0 g of sodium acetate with vigorous stirring. The resulting yellowish-colored mixture was further stirred until a solid precipitate was formed (5-10 min). The reaction mixture was then diluted with cold water (200 mL), the solid product was collected by suction filtration, washed several times with cold water, dried, and recrystallized from ethanol.

$C_{18}H_{13}ClN_2O_3$
MW: 340.92

Yield = 37.13 g (100 %), mp = 271-272 °C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.64 (s, 3H, C*H*3), 6.79 (s, 1H, H-3), 7.20 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.39 (d, *J* = 2.0 Hz, 1H, H-8),7.54 (m, 3H, H-3'+ H-4'+ H-5'), 7.90- 7.93 (m, 2H, H-2'+ H-6'), 8.22 (d, *J* = 8.53 Hz, 1H, H-5), 8.64 (s, 1H, N-H). ^{13}C-NMR (75 MHz, DMSO): δ 26.1 (*C*H3), 102.6 (C-8), 107.4 (C-3), 113.8 (C-6), 118.8 (C-4a), 126.2 (C-1'), 126.8 (C-2'/ C-6'), 127.0 (C-4'), 129.6 (C-3'/ C-5'), 131.7 (C-7), 132.2 (C-5), 148.0 (-C=N), 157.5 (C-8a), 162.6 (C-2), 176.8 (C-4), 188.7 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{18}H_{13}ClN_2O_3Na$ $[M + Na]^+$ 363.05124; found 363.05069.

3.2.2. General procedure of synthesis compounds (79a-n)

To a cold suspension (0 to -10°C) of 1.47 mmol (0.5g) compound (**78)** in 20.0 mL of DMF was added, with stirring, a solution of the appropriate secondary amine and triethylamine (3 mL) in 10 mL of DMF. Stirring was continued at 0 to 5°C for 2-4 h, and then at ambient temperature for 10-12 h. The resulting crude solid product was collected by adding water, washed with water, dried and purified on preparative silica gel TLC plates. Using the same general procedure, the following compounds were prepared:

7-[(2-(2-oxo-1-piperidin-1-ylpropylidene)hydrazino]-2-phenyl-4*H*-chromen-4-one **(79a)**

$C_{23}H_{23}N_3O_3$
MW: 389.45

Yield = 0.43 g (75.7%), mp = 172-173 °C .^{1}H-NMR (300 MHz, $CDCl_3$): δ 1.62 (m, 6H, H_2-3"+ H_2-4"+ H_2-5"), 2.45 (s, 3H, C*H*3), 3.00 (m, 4H, H_2-2"+ H_2-6"), 6.73 (s, 1H, H-3), 7.10 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.32 (d, *J* = 2.0 Hz, 1H, H-8), 7.49-7.51 (m, 3H, H-3'+ H-4'+ H-5'), 7.88-7.91 (m, 2H, H-2'+ H-6'), 8.12 (d, *J* = 8.7 Hz, 1H, H-5), 9.28 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 24.0 (*C*H_3), 26.2 (C-4"), 26.7 (C-3" / C-5"), 49.3 (C-2"/ C-6"), 100.9 (C-8), 107.6 (C-3), 112.6 (C-6), 118.3 (C-4a), 126.2 (C-2'/ C-6'), 127.3 (C-4'), 129.0 (C-3'/ C-5'), 131.5 (C-5), 131.9 (C-1'), 146.7 (C-7), 147.5 (-C=N), 158.0 (C- 8a), 163.0 (C-2), 177.8 (C-4), 195.4 (O=*C*-Me). HRMS(ESI) *m/z*: Calcd for $C_{23}H_{24}N_3O_3$ $[M + H]^+$ 390.18177; found 390.18122.

7-[2-(1-morpholin-4-yl-2-oxopropylidene)hydrazino]-2-phenyl-4*H*-chromen-4-one **(79b)**

$C_{22}H_{21}N_3O_4$
MW: 391.42

Yield = 0.30 g (51.9 %), mp = 209-210 °C . ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.47 (s, 3H, CH_3), 3.08 (t, J = 4.5 Hz, 4H, H_2-2"+ H_2-6"), 3.8 (t, J = 4.5 Hz, 4H, H_2-3"+ H_2-5"), 6.72 (s, 1H, H-3), 7.13 (dd, J = 8.7, 2.0 Hz, 1H, H-6), 7.34 (d, J = 2.0 Hz, 1H, H-8), 7.46-7.50 (m, 3H, H-3'+ H-4'+ H-5'), 7.87-7.90 (m, 2H, H-2'+ H-6'), 8.12 (d, J = 8.7 Hz, 1H, H-5), 9.41 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 26.1 (CH$_3$), 48.3 (C-3"/ C-5"), 67.4 (C-2" / C-6"), 101.2 (C-8), 107.6 (C-3), 112.7 (C-6), 118.6 (C-4a), 126.2 (C-2'/ C-6'), 127.4 (C-4'), 129.1 (C-3'/ C-5'), 131.6 (C-5), 131.8 (C-1'), 144.7 (C-7), 147.2 (-C=N), 157.9 (C-8a), 163.1 (C-2), 177.7 (C-4), 195.1 (O=C-Me). HRMS (ESI) *m/z*: Calcd for $C_{22}H_{20}N_3O_4$ $[M - H]^-$ 390.14538 found 390.14593.

7-[2-(2-oxo-1-thiomorpholin-4-ylpropylidene)hydrazino]-2-phenyl-4*H*-chromen-4-one **(79c)**

$C_{22}H_{21}N_3O_3S$
MW: 407.49

Yield = 0.39 g (65.6 %), mp = 217-218 °C. 1H-NMR (300 MHz, $CDCl_3$): δ 2.46 (s, 3H, CH_3), 2.75 (m, 4H, H_2-3"+ H_2-5"), 3.27 (m, 4H, H_2-2"+ H_2-6"), 6.74 (s, 1H, H-3), 7.13 (dd, J = 8.7, 2.0 Hz, 1H, H-6), 7.33 (d, J = 2.0 Hz, 1H, H-8), 7.47-7.51 (m, 3H, H-3'+ H-4'+ H-5'), 7.88-7.91 (m, 2H, H-2'+ H-6'), 8.13 (d, J = 8.7 Hz, 1H, H-5), 9.24 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 26.0 (CH_3), 28.5 (C-3"/ C-5"), 50.4 (C-2"/ C-6"), 101.2 (C-8), 107.6 (C-3), 112.7 (C-6), 118.6 (C-4a), 126.2 (C-2'/ C-6'), 127.4 (C-4'), 129.0 (C-3'/ C-5'), 131.5 (C-5), 131.8 (C-1'), 145.8 (C-7), 147.2 (-C=N), 157.9 (C- 8a), 163.1 (C-2), 177.7 (C-4), 195.1 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{22}H_{22}N_3O_3S$ [M + H]$^+$ 408.13819; found 408.13764.

7-[2-(2-oxo-1-piperazin-1-ylpropylidene)hydrazino]-2-phenyl-4*H*-chromen-4-one**(79d)**

O
4
5
4a
3
6
O
N
7
2
2'
8a
1'
Me
N
H
8
O
3'
N
6'
4'
6''
2''
5'
$C_{22}H_{22}N_4O_3$
MW: 390.44
5''
3''
N
H

Yield = 0.19 g, (32.9 %), mp = 200-202° C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 1.76 (s, 1H, N(4'')-H), 2.46 (s, 3H, CH_3), 2.98 (m, 4H, H_2-2''+ H_2-6''), 3.00 (m, 4H, H_2-3''+ H_2-5''),6.74 (s, 1H, H-3), 7.11 (dd, *J* = 8.7, 1.7 Hz, 1H, H-6), 7.33 (d, *J* = 1.7 Hz, 1H, H-8), 7.49–7.51 (m, 3H, H-3'+ H-5' +H-4'), 7.88–7.91 (m, 2H, H-2'+ H-6'), 8.13 (d, *J* = 8.7 Hz, 1H, H-5), 9.37 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 26.1 (*C*H_3), 46.6 (C-2''/ C-6''), 49.3 (C-3''/ C-5''), 101.1 (C-8), 107.6 (C-3), 112.7 (C-6), 118.5 (C-4a), 126.2 (C-2'/ C-6'), 127.4 (C-4'), 129.1 (C-3'/ C-5'), 131.5 (C-5), 131.9 (C-1'), 145.5 (C-7), 147.3 (-C=N), 158.0 (C- 8a), 163.0 (C-2), 177.8 (C-4), 195.2 (O=*C*-Me).HRMS (ESI) *m/z*: Calcd for $C_{22}H_{23}N_4O_3$ $[M + H]^+$ 391.17702 ; found391.17647.

7-{2-[1-(4-methylpiperazin-1-yl)-2-oxopropylidene]hydrazino}-2-phenyl-4*H*-chromen-4-one **(79e)**

$C_{23}H_{24}N_4O_3$
MW: 404.46

Yield = 0.11 g (17.7 %), mp = 145-147 ^{o}C. ^{1}H-NMR (300 MHz, CDCl$_3$): δ 2.35 (s, 3H, C*H*$_3$), 2.47 (s, 3H, CH$_3$-N), 2.53 (m, 4H, H$_2$-3''+ H$_2$-5''), 3.10 (m, 4H, H$_2$-2''+ H$_2$-6''), 6.76 (s, 1H, H-3), 7.12 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.35 (d, *J* = 2.0 Hz, 1H, H-8), 7.49 (m,1 H, H-4'), 7.50–7.54 (m, 2H, H-3'+ H-5'), 7.90–7.94 (m, 2H, H-2'+ H-6'), 8.15 (d, *J* = 8.7 Hz, 1H, H-5), 9.26 (s, 1H, N-H). ^{13}C-NMR (75 MHz, CDCl$_3$): δ 26.1 (C=O-*C*H$_3$), 46.5 (N-CH$_3$), 48.0 (C-2''/ C-6''), 55.8 (C-3''/ C-5''), 101.0 (C-8), 107.6 (C-3), 112.6 (C-6), 118.5 (C-4a), 126.3 (C-2'/ C-6'), 127.4 (C-4'), 129.1 (C-3'/ C-5'), 131.5 (C-5), 131.9 (C-1'), 145.5 (C-7), 147.3 (-C=N), 158.0 (C- 8a), 163.1 (C-2), 177.8 (C-4), 195.1 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{23}H_{25}N_4O_3$ [M + H]$^{+}$ 405.19267; found 405.19212.

7-{2-[1-(4-ethylpiperazin-1-yl)-2-oxopropylidene]hydrazino}-2-phenyl-4*H*-chromen-4-one **(79f)**

$C_{24}H_{26}N_4O_3$
MW: 418.49

Yield = 0.20 g (32.1 %), mp = 165-167 ^{o}C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 1.08 (t, *J* = 7.2 Hz, 3H, C*H*$_3$-CH$_2$-), 2.38 (s, 3H,C=O-C*H*$_3$), 2.44 (q, *J* = 7.2 Hz, 2H, CH$_3$-C*H*$_2$-N), 2.52 (m, 4H, H$_2$-3"+ H$_2$-5"), 3.08(m, 4H, H$_2$-2"+ H$_2$-6"), 6.69 (s, 1H, H-3), 7.08 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.28 (d, *J* = 2.0 Hz, 1H, H-8), 7.43–7.47 (m, 3H, H-3'+ H-4'+ H-5'), 7.83 – 7.87 (m, 2H, H-2'+ H-6'), 8.08 (d, *J* = 8.7 Hz, 1H, H-5), 9.26 (s, 1H, N-H). 13C-NMR (75 MHz, $CDCl_3$): δ 12.0 (*C*H$_3$-CH$_2$), 26.1 (C=O-*C*H$_3$), 48.0 (N-*C*H$_2$), 49.5 (C-2"/C-6"), 52.5 (C-3"/C-5"), 101.0 (C-8), 107.5 (C-3), 112.7 (C-6), 118.4 (C-4a), 126.2 (C-2'/C-6'), 127.3 (C-4'), 128.5 (C-3'/C-5'), 131.5 (C-5), 131.8 (C-1'), 145.5 (C-7), 147.4 (-C=N), 157.9 (C- 8a), 163.0 (C-2), 177.7 (C-4), 195.1 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{24}H_{27}N_4O_3$ $[M + H]^+$ 419.20832; found 419.20777.

7-{2-[1-(4-benzylpiperazin-1-yl)-2-oxopropylidene]hydrazino}-2-phenyl-4*H*-chromen-4-one **(79g)**

$C_{29}H_{28}N_4O_3$
MW: 480.56

Yield = 0.70 g (99.4 %), mp = 194-195 °C. [1]H-NMR (300 MHz, $CDCl_3$): δ 2.47 (s, 3H, CH_3), 2.56 (m, 4H, H_2-3''+ H_2-5''), 3.09 (m, 4H, H_2-2''+ H_2-6''), 3.57 (s, 2H, N-CH_2-), 6.75 (s, 1H, H-3), 7.11 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.24- 7.34 (m, 5H, H-2'''+ H-3'''+ H-4'''+ H-5'''+ H-6'''), 7.36 (d, J = 2.2 Hz, 1H, H-8), 7.49 (m, 1H, H-4'), 7.50-7.52 (m, 2H, H-3'+ H-5'), 7.90- 7.93 (m, 2H, H-2'+ H-6'), 8.14 (d, *J* = 8.7 Hz, 1H, H-5), 9.28 (s, 1H, N-H). [13]C-NMR (75 MHz, $CDCl_3$): δ 26.2 (*C*H_3), 48.0 (C-2''/C-6''), 53.8 (C-3''/C-5''), 63.2 (CH_2-N), 101.0 (C-8), 107.6 (C-3), 112.7 (C-6), 118.5 (C-4a), 126.3 (C-2'/C-6'), 127.3 (C-4'), 127.4 (C-4'''), 128.4 (C-3'''/ C-5'''), 129.1 (C-3'/C-5'), 129.2 (C-2'''/C-6'''), 131.5 (C-5), 131.9(C-1'), 137.9 (C-1'''), 145.6 (C-7), 147.4 (-C=N), 158.0 (C- 8a), 163.1 (C-2), 177.8 (C-4), 195.2 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{29}H_{29}N_4O_3$ $[M + H]^+$ 481.22397; found 481.22342.

7-{2-[2-oxo-1-(4-pyrimidin-2-ylpiperazin-1-yl)propylidene]hydrazino}-2-phenyl-4*H*-chromen-4-one **(79h)**

$C_{26}H_{24}N_6O_3$
MW: 468.19

Yield = 0.47 g (68.2 %), mp = 202-203 °C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.48 (s, 3H, C*H*$_3$), 3.13 (m, 4H, H_2-3''+ H_2-5''), 3.94 (m, 4H, H_2-2''+ H_2-6''), 6.51(t, *J*= 4.7 Hz, 1H, H-5'''), 6.76 (s, 1H, H-3), 7.15 (d, *J* = 8.6 Hz, 1H, H-6), 7.37 (s, 1H, H-8), 7.50-7.52 (m, 3H, H-3'+ H-4'+ H-5'), 7.90- 7.92 (m, 2H, H-2'+ H-6'), 8.15 (d, *J* = 8.6 Hz, 1H, H-5), 8.32 (d, *J* = 4.7 Hz, 2H, H-4'''+ H-6'''), 9.45 (s, 1H, N-H). ^{13}C-NMR (75 MHz, DMSO): δ 26.7 (*C*H$_3$), 43.9 (C-2''/C-6''), 47.7 (C-3'' + C-5''), 101.6 (C-8), 107.3 (C-3), 110.6 (C-5'''), 113.7 (C-6), 117.7 (C-4a), 126.7 (C-2'/C-4'/C-6'), 129.7 (C-3'/C-5'), 131.8 (C-1'), 132.1 (C-5), 145.1 (C-7), 149.0 (-C=N), 157.7 (C- 8a), 158.5 (C-'''/C-6'''), 161.8 (C-2''), 162.5 (C-2), 176.8 (C-4), 195.7 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{26}H_{25}N_6O_3$ $[M + H]^+$ 469.19881; found 469.19827.

7-{2-[2-oxo-1-(4-phenylpiperazin-1-yl)propylidene]hydrazino}-2-phenyl-4*H*-chromen-4-one **(79i)**

O
4
5
4a
3
O
6
2
2'
N
7
3'
8a
1'
Me
N
O
H
8
N
6'
4'
6''
2''
5'
5''
3''
$C_{28}H_{26}N_4O_3$
MW: 466.53
N
1'''
6'''
2'''
5'''
3'''
4'''

Yield = 0.38 g (54.7 %), mp = 212-213 °C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.50 (s, 3H, CH_3), 3.18 (m, 4H, H_2-3"+ H_2-5"), 3.24 (m, 4H, H_2-2"+ H_2-6"), 6.73 (s, 1H, H-3), 6.89 (t, *J*=4.3 Hz, 1H, H-4'''), 6.95 (d, *J*=7.9 Hz, 2H, H-2'''+ H-6'''), 7.12 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.27 (m, 2H, H-3'''+ H-5'''), 7.34 (d, *J* = 2.0 Hz, 1H, H-8), 7.49 - 7.51 (m, 3H, H-3'+ H-4'+ H-5'), 7.87 – 7.90 (m, 2H, H-2'+ H-6'), 8.13 (d, *J* = 8.7 Hz, 1H, H-5), 9.38 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 26.1 (CH$_3$), 48.2 (C-2"/C-6"), 50.1 (C-3"/C-5"), 101.2 (C-8), 107.6 (C-3), 112.7 (C-6), 116.5 (C-2'''/C-6'''), 118.6 (C-4a), 120.3 (C-4'''), 126.2 (C-2'/C-6'), 127.4 (C-4'), 129.1 (C-3'/C-5'), 129.3 (C-3'''/C-5'''), 131.5 (C-5), 131.7(C-l'), 145.2 (C-1'''), 147.3 (C-7), 151.3 (-C=N), 158.0 (C- 8a), 163.0 (C-2), 177.7 (C-4), 195.2 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{28}H_{27}N_4O_3$ $[M + H]^+$ 467.20832; found 467.20777.

7-(2-{1-[4-(4-methoxyphenyl)piperazin-1-yl]-2-oxopropylidene}hydrazino)-2-phenyl-4*H*-chromen-4-one **(79j)**

$C_{29}H_{28}N_4O_4$
MW: 496.56

Yield = 0.23 g (31.6 %), mp = 207-208 °C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.50 (s, 3H, CH_3), 3.18 (m, 4H, H_2-3"+ H_2-5"), 3.24 (m, 4H, H_2-2"+ H_2-6"), 3.76 (s, 3H, OCH_3), 6.75 (s, 1H, H-3), 6.85 (m, 2H, H-2'''+ H-6'''), 6.93 (m, 2H, H-3'''+ H-5'''), 7.12 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.35 (d, *J* = 2.0 Hz, 1H, H-8), 7.48 – 7.52 (m, 3H, H-3'+ H-4'+ H-5'), 7.89 – 7.92 (m, 2H, H-2'+ H-6'), 8.14 (d, *J* = 8.7 Hz, 1H, H-5), 9.36 (s, 1H, N-H). ^{13}C-NMR (75 MHz, DMSO): δ 26.8 (*C*H_3), 48.0 (C-2"/C-6"), 50.1 (C-3"/C-5"), 55.7 (OMe), 101.5 (C-8), 107.4 (C-3), 113.6 (C-6), 114.8 (C-3'''/C-5'''), 117.6 (C-4a), 118.1 (C-2'''/C-6'''), 126.7 (C-2'/C-6'), 128.8 (C-4'), 129.6 (C-3'/C-5'), 131.1 (C-1'), 132.1 (C-5), 145.3 (C-1'''), 146.2 (C-7), 149.0 (C-4'''), 153.4 (-C=N), 157.7 (C- 8a), 162.5 (C-2), 176.8 (C-4), 195.6 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{29}H_{29}N_4O_4$ $[M + H]^+$ 497.21888; found 497.21833.

7-(2-{1-[4-(2-fluorophenyl) piperazin-1-yl]-2-oxopropylidene}hydrazino)-2-phenyl-4*H*-chromen-4-one **(79k)**

$C_{28}H_{25}FN_4O_3$
MW: 484.52

Yield = 0.21 g (29.1 %), mp = 225-227 °C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.50 (s, 3H, C*H*$_3$), 3.22 (m, 4H, H$_2$-3''+ H$_2$-5''), 3.23 (m, 4H, H$_2$-2''+ H$_2$-6''), 6.76 (s, 1H, H-3), 6.96 – 7.08 (m, 4H, H-3'''+ H-4''', H-5'''+ H-6'''), 7.14 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.37 (d, *J* = 2.0 Hz, 1H, H-8), 7.50 – 7.53 (m, 3H, H-3'+ H-4'+ H-5'), 7.90 – 7.93 (m, 2H, H-2'+ H-6'), 8.15 (d, *J* = 8.7 Hz, 1H, H-5), 9.36 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 26.1 (*C*H$_3$), 48.3 (C-2''/C-6''), 51.4 (C-3''/C-5''), 101.2 (C-8), 107.6 (C-3), 112.7 (C-6), 116.4 (d, ^2JC-F = 20.6 Hz, C-3'''), 118.6 (C- 4a), 119.2 (d, ^4JC-F = 2.9 Hz, C-5'''), 123.0 (d, ^3JC-F = 7.9 Hz, C-4'''), 124.5 (d, ^3JC-F = 3.5 Hz, C-6'''), 126.3 (C-2'/ C-6'), 127.4 (C-4'), 129.1 (C-3'/C-5'), 131.6 (C-5), 131.9 (C-1'), 139.5 (d, ^2JC-F = 8.6 Hz, C-1'''), 145.3 (C-7), 147.3 (-C=N), 155.5 (d, ^1JC-F = 244.6 Hz, C-2'''), 158.0 (C- 8a), 163.1 (C-2), 177.8 (C-4), 195.1 (O=*C*-Me). HRMS(ESI) *m/z*: Calcd for $C_{28}H_{26}FN_4O_3$ $[M + H]^+$ 485.19442; found 485.19835.

7-(2-{1-[4-(4-fluorophenyl)piperazin-1-yl]-2-oxopropylidne}hydrazino)-2-phenyl-4*H*-chromen-4-one **(79l)**

$C_{28}H_{25}FN_4O_3$
MW: 484.52

Yield = 0.18 g (25.3 %), mp = 225-227 °C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.50 (s, 3H, CH_3), 3.22 (m, 4H, H_2-3''+ H_2-5''), 3.25 (m, 4H, H_2-2''+ H_2-6''), 6.75 (s, 1H, H-3), 6.88 – 6.93 (m, 2H, H-2'''+ H-6'''), 6.94-7.00 (m, 2H, H-3'''+ H-5'''), 7.13 (dd, *J* = 8.7, 2.0 Hz, 1H, H-6), 7.35 (d, *J* = 2.0 Hz, 1H, H-8), 7.48- 7.52 (m, 3H, H-3'+ H-4'+ H-5'), 7.88-7.92 (m, 2H, H-2'+ H-6'), 8.14 (d, *J* = 8.7 Hz, 1H, H-5), 9.36 (s, 1H, N-H). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 26.1 (*C*H_3), 48.2 (C-2''/C-6''), 51.1 (C-3''/C-5''), 101.2 (C-8), 107.6 (C-3), 112.6 (C-6), 115.4 (d, ^2JC-F = 22.0 Hz, C-3''' / C-5'''), 118.3 (d, ^3JC-F = 7.6 Hz, C-2''' / C-6'''), 118.6 (C- 4a), 126.2 (C-2'/C-6'), 127.4 (C-4'), 129.1 (C-3'/C-5'), 131.6 (C-5), 131.9 (C-1'), 145.2 (C-7), 147.3 (-C=N), 147.9 (d, ^4JC-F = 2.3 Hz, C-1'''), 157.5 (d, ^1JC-F = 240 Hz, C-4'''), 158.0 (C-8a), 163.1 (C-2), 177.7 (C-4), 195.2 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{28}H_{26}FN_4O_3$ $[M + H]^+$ 485.19442; found 485.19835.

Ethyl-4-[2-oxo-N-(4-oxo-2-phenyl-4*H*-chromen-7-yl)propanehydrazonoyl] piperazine-1-carboxylate **(79m)**

$C_{25}H_{26}N_4O_5$
MW: 462.50

Yield = 0.43 g (63.3 %), mp = 165-166 °C. ^{1}H-NMR (300 MHz, DMSO-d$_6$): δ 1.17 (t, *J* = 7.0 Hz, 3H, C*H*$_3$-CH$_2$), 2.39 (s, 3H, O=C-C*H*$_3$), 2.91 (m, 4H, H$_2$-3''+ H$_2$-5''), 3.53 (m, 4H, H$_2$-2''+ H$_2$-6''), 4.03 (d, *J* = 7.0 Hz, 2H, MeC*H*2), 6.91 (s, 1H, H-3), 7.50-7.57 (m, 5H, H-3'+ H-4'+ H-5'+ H-6 + H-8), 7.94 (d, *J* = 8.7 Hz, 1H, H-5), 8.03 – 8.06 (m, 2H, H-6'+ H-2'), 10.25 (s, 1H, N-H). ^{13}C-NMR (75 MHz, DMSO): δ 15.11 (*C*H$_3$CH$_2$-), 26.6 (O=C-*C*H$_3$), 44.0 (C-2''/C-6''), 47.8 (C-3''/C-5''), 61.3 (Me*C*H$_2$-), 101.6 (C-8), 107.4 (C-3), 113.6 (C-6), 117.7 (C-4a), 126.7 (C-2'/C-4'/C-6'), 129.6 (C-3'/ C-5'), 131.8 (C-1'), 132.1 (C-5), 144.8 (C-7), 148.9 (-C=N), 155.2 (C-8a), 157.7 (O=*C*-N), 162.5 (C-2), 176.8 (C-4), 195.6 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{25}H_{27}N_4O_5$ $[M + H]^+$ 463.19814; found 463.19760.

7-(2-{1-[4-(4-chlorophenyl)piperazin-1-yl]-2-oxopropylidene}hydrazino)-2-phenyl-4*H*-chromen-4-one **(79n)**

$C_{28}H_{25}N_4O_3Cl$
MW: 500.98

Yield = 0.21 g (28.8 %), mp = 204-205 °C. ^{1}H-NMR (300 MHz, DMSO-d_6): δ 2.41 (s, 3H, CH_3), 3.13 (m, 4H, H_2-3''+ H_2-5''), 3.27 (m, 4H, H_2-2''+ H_2-6''), 6.90 (s, 1H, H-3), 6.97 (d, *J*=9.0 Hz, 2H, H-2'''+ H-6'''), 7.23 (d, *J* = 9.0, 2H, H-3'''+ H-5'''), 7.48 – 7.59 (m, 5H, H-3'+ H-4'+ H-5'+H-6 +H-8), 7.94 (d, *J* = 8.7 Hz, 1H, H-5), 8.05 (m, 2H, H-6' + H-2'), 10.2 (s, 1H, N-H). ^{13}C-NMR (75 MHz, DMSO): δ 26.7 (CH_3), 47.7 (C-2''/C-6''), 48.6 (C-3''/ C-5''), 101.6 (C-8), 107.4 (C-3), 113.6 (C-6), 117.5 (C-2'''/C-6'''), 117.7 (C-4a), 122.8 (C-3'''/C-5'''), 126.7 (C-2'/C-6'), 129.2 (C-4'), 129.7 (C-3'/C-5'), 131.9 (C-1'), 132.1 (C-5), 141.4 (C-4'''), 145.1 (C-1'''), 149.0 (C-7), 150.5 (-C=N), 157.7 (C- 8a), 162.6 (C-2), 176.8 (C-4), 195.6 (O=*C*-Me). HRMS(ESI) *m/z*: Calcd for $C_{28}H_{25}N_4O_3Cl$ $[M - H]^-$ 499.15369; found499.15424.

3.2.3. General procedure of synthesis compounds ((80a-k)-81)

To a cold suspension (0 to -10 °C) of 1.47 mmol (0.5 g) compound **(78)** in 20.0 mL of ethanol was added, with stirring, a solution of L-(α)- amino acid methyl esters (2.0 mmol) and triethylamine (3 mL) in 10 mL of ethanol. Stirring was continued at 0 to 5 °C for 2-4 h, and then at ambient temperature for 24 h. The solvent was evaporated and the residue was treated with water (15 mL). The resulting crude solid product was collected by suction filtration, washed with water, dried and purified on preparative silica gel TLC plates. Using the same general procedure, the following compounds were prepared:

3-acetyl-4,5-dihydro-5-isobutyl-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80a)**

$C_{24}H_{23}N_3O_4$
MW: 417.46

Yield = 0.125 g (20 %), mp = 135-136 °C. [α] $_D$ = -120 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 0.98 (dd, *J* = 7.6, 6.2 Hz, 6H, (CH_3)$_2$CH), 1.67 (m, 1H, C*H*Me$_2$), 1.80 (m, 2H, C*H*$_2$-5), 2.54 (s, 3H, O=C-C*H*$_3$), 4.23 (m, 1H, H-5), 5.99 (s, 1H, NH-4), 6.81 (s, 1H, H-3'), 7.51-7.52 (m, 3H, H-3''+ H-4''+ H-5''), 7.78 (dd, *J* = 8.8, 1.8 Hz, 1H, H-6'), 7.90-7.93 (m, 2H, H-2''+ H-6''), 8.01 (d, *J* = 1.8 Hz, 1H, H-8'), 8.23 (d, *J* = 8.8 Hz, 1H, H-5').^{13}C-NMR (75 MHz, $CDCl_3$): δ 21.6 ((*C*H$_3$)$_2$CH), 23.1 (*C*H -Me$_2$), 24.1 (O=C-*C*H$_3$), 42.0 (*C*H$_2$-5), 52.2 (C-5), 107.7 (C-8'), 112.8 (C-3'), 120.7 (C-6'), 121.7 (C-4'a), 126.0 (C-4''), 126.4 (C-2''/C-6''), 129.1 (C-3''/C-5''), 131.7 (C-1''), 131.8 (C-5'), 142.2 (C-7'), 145.0 (C-3), 156.0 (C-8'a), 162.8 (C-2'), 163.8 (C-6), 178.0 (C-4'), 192.9 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{24}H_{22}N_3O_4$ [M - H]$^-$ 416.16103; found 416.16158.

3-acetyl-4,5-dihydro-5-methyl-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80b**)

$C_{21}H_{17}N_3O_4$
MW: 375.38

Yield = 0.41 g (74.6 %), mp = 277-278 °C. [α] $_D$ = - 99 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 1.55 (d, *J* = 6.7 Hz, 3H, C*H*$_3$(CH)-5), 2.54 (s, 3H, O=C-C*H*$_3$), 4.30 (q, *J* = 6.6 Hz, 1H, H-5), 5.93 (s, 1H, NH-4), 6.82 (s, 1H, H-3'), 7.49-7.53 (m, 3H, H-3''+ H-4''+ H-5''), 7.79 (dd, *J* = 8.8, 1.8 Hz, 1H, H-6'), 7.91-7.94 (m, 2H, H-2''+ H-6''), 8.02 (d, *J* = 1.8 Hz, 1H, H-8'), 8.24 (d, *J* = 8.8 Hz, 1H, H-5'). ^{13}C-NMR (75 MHz, DMSO): δ 19.4 (*C*H$_3$(CH)-5), 24.9 (O=C-*C*H$_3$), 49.4 (C-5), 107.6 (C-8'), 113.0 (C-3'), 121.3 (C-6'), 121.4 (C-4'a), 125.5 (C-4''), 126.9 (C-2'' /C-6''), 129.6 (C-3''/C-5''), 131.6 (C-1''), 132.3 (C-5'), 143.6 (C-7'), 145.6 (C-3), 156.1 (C-8'a), 163.4 (C-2'), 163.9 (C-6), 177.0 (C-4'), 193.2 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{21}H_{18}N_3O_4$ [M + H]$^+$ 376.12973; found 376.12918.

3-acetyl-4,5-dihydro-4-methyl-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80c).**

$C_{21}H_{17}N_3O_4$
MW: 375.38

Yield = 0.49g (88.5 %), mp = 181-182 ^{o}C. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.53 (s, 3H, O=C-C*H*$_3$), 3.16 (s, 3H, C*H*$_3$-(N)4), 4.00 (s, 2H, H_2-5), 6.78 (s, 1H, H-3'), 7.47-7.51 (m, 3H, H-3''+ H-4''+ H-5''), 7.80 (dd, *J* = 8.8, 2.0 Hz, 1H, H-6'), 7.88-7.91 (m, 2H, H-2''+ H-6''), 8.04 (d, *J* = 2.00 Hz, 1H, H-8'), 8.20 (d, *J* = 8.8 Hz, 1H, H-5'). ^{13}C-NMR (75 MHz, DMSO): δ 28.3 (O=C-*C*H$_3$), 38.0 (*C*H$_3$-(N)4), 51.8 (C-5), 107.7 (C-8'), 112.3 (C-3'), 120.7 (C-6'), 121.3 (C-4'a), 125.5 (C-4''), 126.9 (C-2''/C-6''), 129.6 (C-3''/C-5''), 131.6 (C-1''), 132.3 (C-5'), 144.9 (C-7'), 145.3 (C-3), 156.1(C-8'a), 160.5 (C-2'), 163.4 (C-6), 177.8 (C-4'), 194.2 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{21}H_{18}N_3O_4$ $[M + H]^{+}$ 376.12973; found 376.12918.

5-((1*H*-indol-3-yl)methyl)-3-acetyl-4,5-dihydro-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80d)**

$C_{29}H_{22}N_4O_4$
MW: 490.51

Yield = 0.20 g (28 %), mp = 136-137 ^oC. [α] $_D$ = - 260 ^{o} (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.29 (s, 3H, O=C-CH_3), 3.31 (m, 2H, CH_2-5), 4.51 (d, *J* = 3.81Hz, 1H, H-5), 6.04 (s, 1H, NH-4), 6.80 (s, 1H, H-3'), 7.02(s, 1H, H-2'''), 7.07 (t, *J* = 7.4Hz, 1H, H-6'''), 7.19 (t, *J* = 7.5 Hz, 1H, H-5'''), 7.39 (d, *J* = 8.1 Hz, 1H, H-7'''), 7.51- 7.57 (m, 5H, H-3''+ H-4''+ H-5''+ H-6'+H-4'''), 7.81 (s, 1H, H-8'), 7.85-7.95 (m, 2H, H-2''+H6''), 8.17 (d, *J* = 8.7 Hz, 1H, H-5'), 8.63 (s, 1H, NH-1''). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 23.9 (*C*H_3), 30.4 (*C*H_2-5), 54.4 (C-5), 107.6 (C-8'), 108.9(C-3'''), 111.5 (C-3'), 113.1 (C-7'''), 118.6(C-4'''), 119.9(C-5'''), 120.9 (C-6'), 121.7 (C-4'a), 122.5 (C-6'''), 123.7(C-2'''), 125.7 (C-4''), 126.4 (C-2''/C-6''), 127.1(C-3'''a), 129.1 (C-3''/C-5''), 131.6 (C-1''), 131.8 (C-5'), 136.5 (C-7'''a), 142.2 (C-7'), 144.9 (C-3), 156.1 (C-8'a), 162.6 (C-2'), 163.8 (C-6), 178.1 (C-4'), 192.7 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{29}H_{21}N_4O_4$ [M - H] $^-$ 489.15628; found 489.15683.

3-acetyl-4,5-dihydro-5-(2-(methylthio)ethyl)-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one **(80e)**

$C_{23}H_{21}N_3O_4S$
MW: 435.50

Yield = 0.24 g (37.6 %), mp = 189-190 ^{o}C. $[\alpha]_D$ = -110 ^{o} (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.10 (s, 3H, CH_3S), 2.17 (m, 2H, CH_2-5), 2.54 (s, 3H, O=C-CH_3), 2.67 (t, *J* = 7.1 Hz, 2H, CH_2S), 4.40 (t, *J* = 5.1 Hz, 1H, H-5), 6.25 (s, 1H, NH-4), 6.81 (s, 1H, H-3'), 7.50- 7.52 (m, 3H, H-3''+ H-4''+ H-5''), 7.77 (dd, *J* = 8.8, 2.0 Hz, 1H, H-6'), 7.90 – 7.93 (m, 2H, H-2''+ H-6''), 8.0 (d, *J* = 2.0 Hz, 1H, H-8'), 8.23 (d, *J* = 8.8 Hz, 1H, H-5'). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 15.4 (CH_3S), 24.1 (O=CCH_3), 29.7(CH_2S), 32.0 (CH_2CH), 53.0 (C-5), 107.7 (C-8'), 112.9 (C-3'), 120.7 (C-6'), 121.8 (C-4'a), 125.9 (C-4''), 126.4 (C-2''/C-6''), 129.1 (C-3''/C-5''), 131.67 (C-1''), 131.74 (C-5'), 142.1 (C-7'), 144.9 (C-3), 156.2 (C-8'a), 162.1 (C-2'), 163.8 (C-6), 177.9 (C-4'), 192.7 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{23}H_{22}N_3O_4S$ $[M + H]^+$ 436.1331; found 436.13255.

3-acetyl-5-benzyl-4,5-dihydro-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80f**)

Yield = 0.29 g (44.3 %), mp = 151-152 °C. [α] $_D$ = - 350 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.41 (s, 3H, CH_3), 2.63 (m, 2H, -CH_2Ph), 4.48 (m, 1H, H-5), 5.92 (s, 1H, NH-4), 6.82 (s, 1H, H-3'), 7.13 -7.16 (m, 2H, H-2'''+ H-6'''), 7.28 -7.32 (m, 3H, H-3'''+ H-4'''+ H-5'''), 7.50 – 7.54 (m, 3H, H-3''+ H-4''+ H-5''), 7.66 (dd, *J* = 8.8, 2.0 Hz, 1H, H-6'), 7.91 – 7.93 (m, 2H, H-2''+ H-6''), 7.94 (d, *J* = 2.0 Hz, 1H, H-8'), 8.23 (d, *J* = 8.8 Hz, 1H, H-5'). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 23.9 (O=C-CH_3), 40.3 (CH_2-Ph), 55.2 (C-5), 107.8 (C-8'), 113.0 (C-3'), 120.8 (C-6'), 121.9 (C-4'a), 125.9 (C-4''), 126.4(C-2''/C-6''), 127.7(C-4'''), 129.0 (C-2'''/C-6'''), 129.1 (C-3''/C-5''), 129.6 (C-3'''/C-5'''), 131.7 (C-1''), 131.8 (C-5'), 134.9 (C-1'''), 141.9 (C-7'), 144.7 (C-3), 156.2 (C-8'a), 161.9 (C-2'), 163.8 (C-6), 177.9 (C-4'), 192.6 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{27}H_{20}N_3O_4$ [M - H]$^-$ 450.14538; found 450.14593.

Methyl3-(3-acetyl-1,4,5,6-tetrahydro-6-oxo-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-5-yl)propanoate (**80g)**

$C_{24}H_{21}N_3O_6$
MW: 447.44

Yield = 0.30 g (45.4 %), mp = 134-135 ^oC. [α] $_D$ = -140 ^{o} (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.23 (m, 2H,CH_2CH), 2.54 (m, 2H, CH_2COOMe), 2.53 (s, 3H, O=C-CH_3), 3.67(s, 3H, CH_3OCO), 4.32 (m, 1H, H-5), 6.25 (s, 1H, NH-4), 6.82 (s, 1H, H-3'), 7.48 – 7.52 (m, 3H, H-3''+ H-4''+ H-5''), 7.77 (dd, *J* = 8.8, 1.8 Hz, 1H, H-6'), 7.90 – 7.93 (m, 2H, H-2''+ H-6''), 8.00 (d, *J* = 1.8 Hz, 1H, H-8'), 8.22 (d, *J* = 8.8 Hz, 1H, H-5'). ^{13}C-NMR (75 MHz, $CDCl_3$): δ 24.1 (O=C-*C*H_3), 28.6 (*C*H_2CH), 29.7 (*C*H_2COOMe), 52.1 (CO_2*C*H_3), 53.1 (C-5), 107.7 (C-8'), 112.9 (C-3'), 120.7 (C-6'), 121.9 (C-4'a), 125.9 (C-4'') , 126.4 (C-2''/C-6''), 129.1 (C-3''/C-5''), 131.7 (C-1''), 131.8 (C-5'), 142.3 (C-7'), 144.8 (C-3), 156.2 (C-8'a), 161.8 (C-2'), 163.8 (C-6), 173.2 (*C*O_2Me),177.9 (C-4'), 192.6 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{24}H_{22}N_3O_6$ $[M + H]^+$ 448.15086; found 448.15031.

3-acetyl-4, 5-dihydro-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80h**)

Yield = 0.51 g (96.8 %), mp = 275-276 °C. ^{1}H-NMR (300 MHz, DMSO-d$_6$): δ 2.46 (s, 3H, CH_3), 4.04 (s, 2H, H_2-5), 7.02 (s, 1H, H-3'), 7.56-7.58 (m, 3H, H-3"+ H-4"+ H-5"), 7.72 (br. s, 1H, NH-4), 7.85 (dd, *J* = 8.8, 1.7 Hz, 1H, H-6'), 8.04-8.12 (m, 4H, H-5'+ H-8'+H-2"+ H-6"). ^{13}C-NMR (75 MHz, DMSO): δ 24.9 (CH_3), 43.8 (*C*-5), 107.6 (C-8'), 112.8 (C-3'), 121.2 (C-6'), 121.3 (C-4'a), 125.4(C-4"), 126.9 (C-2"/C-6"), 129.7 (C-3"/C-5"), 131.6 (C-1"), 132.4 (C-5'), 143.7 (C-7'), 145.4 (C-3), 156.1 (C-8'a), 160.8 (C-2'), 163.3 (C-6), 177.0 (C-4'), 193.1 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{20}H_{16}N_3O_4$ [M + H]$^+$ 362.11408; found 362.11353.

3-acetyl-4,5-dihydro-5-(hydroxymethyl)-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80i)**

$C_{21}H_{17}N_3O_5$
MW: 391.38

Yield = 0.71 g (100 %), mp = 271-272 °C. [α] D = -226 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, DMSO-d$_6$): δ 2.43 (s, 3H, C*H*$_3$), 3.57-3.85 (m, 2H, CH$_2$-5), 4.17 (br s, 2H, H-5), 5.18 (t, *J* = 5.7 Hz, 1H, OH), 7.02 (s, 1H, H-3'), 7.55-7.57 (m, 3H, H-3''+ H-4''+ H-5''), 7.70 (br. s, 1H, NH-4), 7.83 (dd, *J* = 8.7, 1.7 Hz, 1H, H-6'), 8.05(d, *J* = 8.7 Hz, 1H, H-5'), 8.09 (d, *J* = 1.7 Hz, 1H, H-8'), 8.11(m, 2H, H-2''+ H-6''). ^{13}C-NMR (75 MHz, DMSO): δ 24.8 (*C*H$_3$), 56.7 (*C*-5), 64.0 (CH$_2$-5), 107.5 (C-8'), 112.9 (C-3'), 121.2 (C-6'), 121.3 (C-4'a), 125.4 (C-4''), 126.9 (C-2''/C-6''), 129.6 (C-3''/C-5''), 131.6 (C-1''), 132.4 (C-5'), 143.4 (C-7'), 145.6 (C-3), 156.1 (C-8'a), 162.1 (C-2'), 163.3 (C-6), 177.0 (C-4'), 193.3 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{21}H_{17}N_3O_5Na$ $[M + Na]^+$ 414.10659; found 414.10604.

3-acetyl-4,5-dihydro-5-(1-hydroxyethyl)-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-6(1*H*)-one (**80j)**

$C_{22}H_{19}N_3O_5$
MW: 405.40

Yield = 0.48 g (80.4 %), mp = 267-268 °C. [α] $_D$ = -150 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, DMSO-d$_6$): δ 1.09 (d, *J* = 6.4 Hz, 3H, CH_3-CH), 2.44 (s, 3H, O=C-CH_3), 3.92 (br s, 1H, H-5), 4.04 (br s,1H, CH-5), 5.02 (d, *J* = 5.5 Hz, 1H, OH), 77.02 (s, 1H, H-3'), 7.55-7.58 (m, 3H, H-3''+ H-4''+ H-5''), 7.62 (br. s, 1H, NH-4),7.83 (dd, *J* = 8.7, 1.7 Hz, 1H, H-6'),), 8.05 (d, *J* = 8.7 Hz, 1H, H-5'), 8.09-8.12(m, 2H, H-8'+ H-2''+ H-6''). ^{13}C-NMR (75 MHz, DMSO): δ 19.6 (CH_3-CH), 24.9 (O=C-CH_3), 59.5 (CH-H_5), 68.8 (*C*-5), 107.6 (C-8'), 113.0 (C-3'), 121.2 (C-6'), 121.4 (C-4'a), 125.4 (C-4''), 126.9 (C-2''/C-6''), 129.7 (C-3''/C-5''), 131.6 (C-1''), 132.4 (C-5'), 143.5 (C-7'), 145.7 (C-3), 156.1 (C-8'a), 162.6 (C-2'), 163.3 (C-6), 177.0 (C-4'), 193.3 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{22}H_{20}N_3O_5$ [M + H]$^+$ 406.1403; found 406.13975.

Methyl2-(3-acetyl-1,4,5,6-tetrahydro-6-oxo-1-(4-oxo-2-phenyl-4*H*-chromen-7-yl)-1,2,4-triazin-5-yl)acetate **(80k).**

Yield = 0.12 g (18.1%), mp = 222-224 °C. [α] $_D$ = -60 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 2.43(s, 3H, CH_3), 2.84-3.05 (m, 2H, H_2-5), 3.37(s, 1H, OMe) , 4.55 (m, 1H, H-5), 7.02 (s, 1H, H-3'),7.52-7.57 (m, 3H, H-3''+ H-4''+ H-5''), 7.78-7.82 (m, 2H, , NH-4+H-6'), 8.04-8.11 (m, 4H, H-5'+ H-8'+H-2''+ H-6''). ^{13}C-NMR (75 MHz, DMSO): δ 24.8 (O=C-*C*H_3), 37.5 (CH_2-5), 50.6 (O=C-OCH_3), 52.2 (*C*-5), 107.6 (C-8'), 113.0 (C-3'), 121.3 (C-6'), 121.4 (C-4'a), 125.4 (C-4''), 126.9 (C-2''/C-6''), 129.6 (C-3''/C-5''), 131.6 (C-1''), 132.3 (C-5'), 142.6 (C-7'), 145.5 (C-3), 156.1 (C-8'a), 162.5 (C-2'), 163.3 (C-6), 171.0 (*C*O_2Me),177.0 (C-4'), 193.2 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{23}H_{20}N_3O_6$ $[M + H]^+$ 434.13521; found 434.13466.

4-acetyl-6,7,8,8a-tetrahydro-2-(4-oxo-2-phenyl-4*H*-chromen-7-yl)pyrrolo[1,2-d][1,2,4]triazin-1(2*H*)-one (**81**)

$C_{23}H_{19}N_3O_4$
MW: 401.41

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Yield = 0.39 g (66.5 %), mp = 210-211 °C. [α] $_D$ = + 404 ° (*c* ~ 1, DMF). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 1.96 (m, 2H, H_2-7), 2.33 (m, 2H, H_2-8), 2.53 (s, 3H, C*H*$_3$), 3.83 (m, 1H, H-8a), 4.00 (m, 2H, H_2-6), 6.79 (s, 1H, H-3'), 7.49 (m, 3H, H-3''+ H-4''+ H-5''), 7.81 (dd, *J* = 8.8, 1.8 Hz, 1H, H-6'), 7.90 (m, 2H, H-2''+ H-6''), 8.04 (d, *J* = 1.8 Hz, 1H, H-8'), 8.21 (d, *J* = 8.8 Hz, 1H, H-5'). ^{13}C-NMR (75 MHz, $CDCl_3$): 24.3 (*C*H_3), 26.4 (C-7), 27.5 (C-8), 50.1 (C-6), 58.0 (C-8a), 107.7 (C-8'), 112.3 (C-3'), 120.3 (C-6'), 121.5 (C-4'a), 125.9 (C-4''), 126.3 (C-2''/C-6''), 129.1 (C-3''/C-5''), 131.6 (C-1''), 131.7 (C-5'), 143.9 (C-7'), 144.8 (C-3), 156.2 (C-8'a), 162.1 (C-2'), 163.7 (C-1), 177.9 (C-4'), 194.3 (O=*C*-Me). HRMS (ESI) *m/z*: Calcd for $C_{23}H_{20}N_3O_4$ $[M + H]^+$ 402.14538; found 402.14483.

4. Results and discussions

4.1. Synthesis of 2-oxo-N-(4-oxo-2-phenyl-4*H*-chromen-7-yl) propane hydrazonyl chloride(78)

The 2-oxo-N-(4-oxo-2-phenyl-4H-chromen-7-yl) propane hydrazonyl chloride (**78**) was prepared *via* diazotization of 7-aminoflavone (**77**), followed by coupling with 3-chloro-2, 4 -pentanedione (Japp-Klingemann reaction) in basic medium (NaOAc or pyridine), as in (Scheme 1). The resulting yellow-colored solid precipitate was collected and recrystallized from acetonitrile.

Scheme 1. Synthesis of N-(4-oxo-2-phenyl-4H-chromen-7-yl)-2-oxopropane hydrazonylchloride.

The Japp-Klingemann reaction is a special case of the coupling of arene diazonium salt with activated methinyl compounds. It is performed by direct electrophilic attack of the diazonium cation on carbanion. The resulting intermediate azo compound is unstable,

undergoing azo to hydrazone conversion through loss of acetyl group as shown in (scheme 2) (Yao and Resnick, 1962).

Scheme 2. Japp-Klingmann reaction mechanism.

The structure of **(78)** is supported by analytical and spectral data (NMR and MS). Thus, the ^{1}H-NMR spectrum showed a singlet signal at 2.64 ppm which is attributed to the acetyl methyl protons. The exchangeable NH proton appears as downfield broad singlet

signal at 8.64 ppm. The spectrum also shows three signals in the aromatic region at 7.20, 7.39 and 8.22 ppm which are assigned to H-6 (dd, J = 8.7, 2.0 Hz), H-8 (d, J = 2.0 Hz) and H-5 (dd, J = 8.53 Hz) proton, respectively. The latter H-6 proton is coupled with H-5 and H-8 protons. Two other broad multiplite signals appear at 7.54, 7.90 – 7.93ppm which are assigned to (H-3'+ H-4' + H-5') and (H-2'+ H-6') protons, respectively. The vinylic H-3 proton appears as singlet at 6.79 ppm. The ^{13}C-NMR spectrum of compound (**78**) revealed clearly that all of different carbon atoms are detectable; the keto group resonates at 188.7 ppm, the methyl carbon (acetyl group) appears at 26.1 ppm. DEPT experiment confirmed the presence of nine C-H atoms in the range 102.6-132.2 ppm.

4.2. Preparation of amidrazones (79a-n)

Amidrazones (**79a-n)** were prepared by reaction of hydrazonoyl chloride (**78**), in presence of triethylamine, with the appropriate secondary amines at 0 °C in DMF. Secondary amines, acting as nucleophiles, add readily to nitrile imines (generated *in situ* from the respective hydrazonoyl chlorides). The interaction between nitrile imines and amines is expected to yield the corresponding Z-amidrazones (Hussein et al., 1984) as the kinetically controlled products (Scheme 3).

Scheme 3. Synthesis of amidrazone (79a-n).

Compounds **79a-n**

entry	a	b	c	d	e	f	g	h	i	j	k	l	m	n
X	CH_2	S	O	NH_2	NMe	NEt	NBz	N(2-pyrimidine)	N(p-Ph)	N(p-C_6H_4OMe)	N(o-C_6H_4F)	N(p-C_6H_4F)	N(CO_2Et)	N(p-C_6H_4Cl)

The structure of **(79e)** is supported by analytical and spectral data (NMR and MS).Thus the ^{1}H-NMR spectrum showed that the N-methyl proton resonates as singlet at 2.47 ppm, The exchangeable NH proton appears as a downfield broad singlet signal around 9.26 ppm, The H-2" + H-6" methylene protons in the piperazine moiety resonate at 3.10 ppm, and are more deshielded than the H-3" + H-5" methylene protons resonating at 2.53 ppm. The ^{13}C-NMR spectrum of compound (**79e**) showed that the N-methyl carbon resonates at 46.5, the methylene carbons in the piperazine moiety (C-2" / C- 6") and (C-3" / C-5") resonate at 48.0 and 55.8 respectively, ^{1}H- and ^{13}C-NMR signal assignments to the various hydrogens and carbons in (**79a-n)**, were deduced in similar basis.

Considering compounds (**79d-n)**, the effect of the group attached to the N (4) atom of the piperazine could be noticed. If the attached group is electron-withdrawing, as in compounds (**79h-n)**, δ-value of piperazine protons will be shifted downfield as compared to the unsubstituted compound at N(4) (**79d**). Conversely, in compounds (**79d-g)** which

have electron-releasing groups through inductive effect, the δ-value of piperazine protons is upfield shifted. ^{13}C-NMR spectrum of (**79k**) reveals clearly the effect of fluorine atom onto the adjacent carbons; thus the following carbons showed through bonds coupling with the fluorine atom (C-2''', ^{1}J = 244.6 Hz), (C-3''', ^{2}J = 20.6 Hz), (C-1''', ^{2}J = 8.6 Hz), (C-4''', ^{3}J = 7.9 Hz), (C-6''', ^{3}J = 3.5 Hz) and(C-5''', ^{4}J = 2.9 Hz). ^{13}C-NMR spectrum of (**79l**) also shows the coupling with the fluorine atom (C-4''', ^{1}J = 145.7 Hz), (C-3'' / C-5'', ^{2}J = 22.0 Hz,), (C-2'' / C-6'', ^{3}J = 7.6 Hz,), and (C-1'', ^{4}J = 2.3 Hz,).

4.3. Preparation of 4, 5-dihydro-1,2,4-triazin-6-one (80a-k)

In the present study, α-amino esters are found to react with hydrazonoyl chlorides (**78**) at low temperature, in the presence of triethyl amine, to give directly the corresponding 4,5-dihydro-1,2,4-triazin-6-ones (Scheme 4). Under such reaction conditions, the hydraznoy1 chlorides (**78**) are assumed to be transformed completely into the respective intermediate nitrile imines, as given in (Scheme 5).

Scheme 4. Synthesis of 4,5-dihydro-1,2,4-triazin-6-ones (80a-k)

Compounds **80a-k**

	R1	R2
a	H	H
b	CH_3	H
c	H	CH_3
d	$^{-}CH_2$	H
e	$CH_2CH_2SCH_3$	H
f	CH_2Ph	H
g	CH_2CO_2Me	H
h	$CH_2CH_2CO_2Me$	H
i	CH_2OH	H
j	$CH(OH)CH_3$	H
k	$CH_2CH(CH_3)_2$	H

The α-amino esters are considered as nitrogen nucleophiles containing a suitably located electrophilic center. α-Amino esters add through the amino group onto nitrile imines (in a similar manner to amines) to give the corresponding Z-amidrazone ester intermediates. The latter acyclic Z-amidrazone adducts are likely to undergo intramolecular cyclization to (**80**) (Scheme 5).

Scheme 5. Mechanism of preperation of 4,5-dihydro[1,2,4]triazin-6-one (80a-k)

The ^{1}H-NMR spectra show that the acetyl protons resonate at 2.17- 2.50 ppm and 2.29-2.54 ppm for (**79a-n**) and (**80a-k**), respectively. The spectrum also show that the proton signal at position 3 appears as a singlet in the range 6.69-6.76 ppm for compounds (**79a-n)**, whereas in compounds **(80a-k),** the corresponding H-3' proton is more deshielded and resonates at 6.71-6.82 ppm. The H-5 proton resonates as a doublet (J = 8.7 Hz) at 8.08-8.22 in (**79a-n)**, while in (**80a-k)** the corresponding H-5' proton resonates at 8.17-8.24 ppm. The H-6 proton in (**79a-l)** resonates as doublet of doublet at 7.08-7.14 ppm (J = 8.7, 2.0 Hz), while the corresponding proton at H-6' belonging to (**80a-k)** is more deshielded and resonates at 7.66-7.81 ppm ($CHCl_3$). The H-8 proton (**79a-l)** and its corresponding proton at H-8' of **(80a-k)** resonates as doublet at 7.28-7.37 and 7.90-8.04 ppm (J = 2.0

Hz), respectively ($CHCl_3$). The H-6 and H-8 proton for compounds **(79m-n)** resonate as multiplit at 7.49-7.58 ppm (DMSO-d_6).The (H-3'+ H-4' + H-5'),(H-2'+ H-6') protone in compounds (**79a-n**) resonate as multiplit at 7.46-7.54, 7.83-7.94 ppm respectively, whereas in compounds **(80a-g),** the corresponding (H-3"+ H-4" + H-5"),(H-2"+ H-6") proton resonates as multiplite at 7.47-7.57 and 7.88-7.94 ppm respectively ($CHCl_3$). In compounds (**80h-k**) the (H-3"+ H-4" + H-5"), (H-2"+ H-6") proton resonates as multiplite at 7.55-7.58 and 8.08-8.12 ppm, respectively (DMSO-d_6).

The N-H proton in (**79a-l)** resonates at 9.24-9.45 ppm($CHCl_3$), in compounds (**79m-n)** N-H proton appears at 10.20-10.25 ppm (DMSO-d_6). While compounds (**80a-k)** are devoid of N-H signal, indicative of cyclization of Z-amidrazone esters. The ^{13}C-NMR spectra of (**79a-n)** show that the C-H carbons of the flavone appear at 107.3-107.6, 131.7-132.1, 112.6-113.7 and 100.9-101.6 ppm for C-3, C-5, C-6 and C-8, respectively. In the ^{13}C-NMR spectra of the cyclized products (**80a-k),** a remarkable difference from the above values could be observed for (**79a-n)** since the C-H carbons (at flavone moiety) appear more deshielded at 112.3-113.0, 131.7-132.4, 120.1-121.3, 107.5-107.8 ppm for C-3', C-5', C-6' and C-8', respectively. The carbonyl carbon of acetyl group appears at 195.1-195.8 and 192.7-194.3 ppm for (**79a-n**) and (**80a-k)**, respectively. On the other hand, the C-4 carbonyl carbon appears at 176.8- 177.8 and 177.0-178.1 ppm for (**79a-n)** and (**80a-k)**. Compounds (**80a-g)** have N-H proton (at 4-position) resonating as singlet in the range 7.70-7.76 ppm ($CHCl_3$), while in compounds (**80h-k)** N-H proton resonates at 3.31-3.27 ppm (DMSO-d_6). The C-5 proton in compounds (**80a-k**) appear as part of ABX system in the range 4.00-4.51 ppm, except for (**80b)** (quartet), (**80c)** and (**80h)** (singlet).

This is due to coupling with the neighboring two diastereotopic protons (of the CH_{2-} methylene group.

4.4. Preparation of pyrrolo [1, 2-d][1,2,4]triazin-1-one (81)

Compound (**81)** was prepared in a similar manner *via* reacting (L)-proline methyl ester with hydrazonoyl chloride (**78**) in presence triethylamine, at 0 °C in ethanol (scheme 18). Proline is a cyclic secondary α-amino acid and reacts with nitrile imine through attack at the carbon of nitrile imine whereas the carboxyl group cyclizes to form the [1, 2, 4]triazinone ring fused onto the pyrrolidine ring, as shown in (scheme 6).

Scheme 6. Synthesis of pyrrolo[1,2-d][1,2,4]triazin-1-one (81)

The assigned structure of compound (**81)** is supported by spectral (NMR and HRMS) data. The ^{1}H and ^{13}C spectra are used for assignment of the different protons and carbons. Thus, the ^{1}H-NMR spectrum shows three different methylene proton's signals at 1.96, 2.33 and 4.00 ppm which are assigned to (H_2-7), (H_2-8) and (H_2-6), respectively. Those protons appear as multiplets due to mutual coupling with each other. Another signal appears as multiplet resonating at 4.00 ppm which is assigned to C-8a proton.The (H-3"+ H-4" + H-5"), (H-2"+ H-6") protones in compounds (**81**) resonate as multiplite at 7.49, 7.90, respectively. The ^{13}C-NMR spectrum of compound (**81)** reveal clearly the presence of three methylene carbons in addition to one aliphatic C-H carbon and nine aromatic C-

H carbons. The ^{13}C-NMR spectrum also indicates the presence of the keto group at 194.3 ppm, while the carbonyl carbon of the (C-4') resonates at 177.9 ppm, and the C-1 carbon appears at 163.7 ppm.

4.5. Antitumor activity

The antitumor activity of the **79a-m** compounds was characterized by conducting cell viability assay using tetrazolium dye 3-(4,5-dimethylthiazol-2-yl)-2,5diphenyltetrazolium bromide (MTT) against MCF7 , T47D breast cancer and K562 leukemia cells. Cell viability was assessed, after 3 days of treatment, with tetrazolium dye 3-(4,5 dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT). For the IC50 determination, the cells were treated with increasing concentrations of the tested compound (1.56–100 uM). IC_{50} concentrations were obtained from the dose–response curves using Graph Pad Prism Software 5 (San Diego California USA, www.graphpad.com).

MCF7 breast cancer

The MCF7 screening test showed that 7 compounds (**79a, 79d, 79e, 79f, 79g, 79h, 79m**) have a significant anti–MCF7activity. Those 7 compounds were able to reduce the viability after 72 hours to less than 50% (Table1.). The IC50 values for the potential compounds against the MCF7 showed that 3 compounds (**79d, 79g, 79h**) have IC50 values less than 10 uM (Table 2.).

K562 leukemia

The K562 leukemia screening test showed that 6 compounds have a significant anti–MCF7 activity. Those 6 (**79a, 79b, 79d, 79e, 79f, 79j**) compounds were able to reduce the viability after 72 hours to less than 50% (Table1.) The IC50 values for the potential

compounds against the K562 showed that 2 compounds (**79a and 79d**) have IC50 values less than 10 uM (Table 2.).

T47D breast cancer

More encouraging results were obtained for the T47D breast cancer screening test. The number of compounds that scored an IC_{50} of less 10 uM increased to 4 (**79a, 79d, 79g, 79h**) and the IC_{50} values were lower than those scored in the MCF7 breast cancer case. The IC_{50} values for 79d and 79g compounds against T47D are showed to be of less than 2 uM (Table 2.).

Compound	MCF7% survival	standard deviation	K562 % survival	standard deviation
79a	**18.01**	0.01	**44.58**	2.71
79b	66.77	6.19	**19.83**	0.02
79c	76.73	7.93	94.56	3.75
79d	**10.28**	0.02	**7.69**	0.07
79e	**7.98**	0.24	**5.18**	0.01
79f	**9.77**	0.46	**15.88**	0.02

79g	**44.32**	0.02	83.27	6.10
79h	**20.27**	0.01	83.90	3.85
79i	75.58	0.01	64.85	0.08
79j	83.70	0.10	**42.19**	0.07
79k	66.42	0.04	72.62	0.11
79l	88.54	0.05	60.86	0.09
79m	**14.36**	0.14	68.27	0.05

Table1. Percentage cell survival of MCF7 and K562 following 72 hours exposure to 50 uM f all compounds.

Compound	IC_{50} T47D (µM)	standard deviation	IC_{50} MCF-7 (µM)	standard deviation	IC_{50} K562 (µM)	standard deviation
Doxorubicin	0.33	0.05	0.31	0.01	1.41	0.31
79a	**8.79**	0.80	21.59	5.87	**2.56**	0.57
79b					20.18	1.41
79d	**1.42**	0.13	**5.91**	1.61	**5.02**	0.78
79e	15.76	1.38	22.37	4.56	16.15	4.17
79f	53.37	4.03	56.79	8.88	35	1.06
79g	**1.92**	0.35	**2.75**	0.73		
79h	**4.31**	0.54	**8.75**	1.38		
79j					14.07	0.52
79m	11.05		13.56	1.82		

Table 2. Effects of compounds that have shown potential activity on the screening assay T47D, MCF-7 and K562. Doxorubicin is used as a Positive control.

5. Conclusion

A new flavone-7-yl hydrazonoyl chloride (**78**) was generated from 7-amino-flavone (**77**) *via* the Japp-Klingemann reaction. In presence of triethylamine, the hydrazonoyl chloride (**78**) reacts with selected set of secondary amines in ethanol at 0 °C to form flavone-7-yl amidrazones (**79**). Under similar conditions, compound (**78**) reacts with L-(α)-amino esters to form flavone-7-yl 4,5-dihydro[1,2,4]-triazin-6-ones (**80**) and pyrrolo[1,2-d][1,2,4]triazin-1-one (**81**). The latter being derived from proline methyl ester. Preliminary antitumer testing results of these compounds showed them to exhibited good to significant antitumor activity against HL-60 and MCF-7 cell lines.

6. References

Abdel-Jalil, R. J., El Momani, E. Q., Hamad, M., Voelter, W., Mubarak, M. S., Smith, B. H. and Peters, D. G. (2010), Synthesis, antitumor activity, and electrochemical behavior of some piperazinyl amidrazones. **Monatsh. Chem.**, 141, 251-258.

Acheson, R. M., (1976), **An Inroduction to the Chemistry of Heterocyclic Compounds**, (3rd ed.), New York, John Wiley and Sons.

Adam, W., Golsch, D., Hadjiarapoglou, L. and patonay, T., (1991), Epoxidation of Flavones by Dimethyldioxirane. **J.org.Chem**., 56, 7292. -7297.

Adam, W., Hadjiarapoglou, L. and levia, A., (1992), **Synthesis**, 436.

Akama, T., lkeda, S. and Shida, Y. (1993), **Eur Patent Appl EP 556 720: Chem Abstract**, 120, 106-766.

Akinaga. S., Gomt, K., Kasai, M., Morimoto, M., Shlda, Y. and Sugava, T., (1990) **Jpn Kokai Tokkyo Koho JP 02 I38 277, Chrm Abe,** 113,1775

Allan, J. and Robinson, R., (1924). **J. Chem. Soc.**, 125, 2192.

Alloway, G., Brossi, A., Kilgore, N., Lee, K. H., Yu, D. and Wild, C., (2003), **Bioorg. Med. Chem. Lett**., 13, 1575.

Altounyan, R. E. and Howell, J. B. (1967). A double-blind trial of disodium cromoglycate in the treatment of allergic bronchial asthma. **Lancet**, 2, 539-542.

Aly, A. A. and Nour-El-Din, A. M.,(2008), Functionality of amidines and amidrazones. **ARKIVOC**, 1, 153-194.

Arimitsu, J., Hagihara, K., Higa, S., Hirano, T., Kawai, Kawase, I., M., Kuwahara, Y., Maruta, M., Ohkawara,T., Ogata, A., Shima, Y., Tanaka, T., and Yamadori, T., (2007), Flavonoids and related compounds as anti-allergic substances. **Allergol. Int**., 56, 23-113.

Artusi, R., Caselli, G., Giordani, A., Lanza, M., Makovec, F., Mennuni, L., Rovati, L. C., Pucci, S.and Verpilio, I., (2009), **2-Aryl And 2-heteroaryl 4H-1-benzopyran-4-one-6-amidino Derivatives For The Treatment Of Arthritis, Cancer And Related Pain**. Patent no. WO 2009/109230 A1.

Azima, M., Brown, B. J., Cutting, W. C., Dreisbach, R. H., Neff, B. J. and Wray, J.,(1951), **Stanford Med. Bull**., 9, 236.

Azqueta, A., Cabrera, M., Castellano, E. E., Cerecetto, H., de Cerain, A. L., Falchi, G., Gonzalez, M. , Lavaggi, M. L., Monge, A., Piro, O. E., Sagrera, G., Seoane, G., Simoens, M. and Vidal, A., (2007), Synthetic chalcones, flavanones, and flavones as antitumoral agents: Biological evaluation and structure-activity relationships**. Bioorg. Med. Chem.**, 15, 3356-3367.

Bahceci, S., Yuksek, H. and Ikizler, A. A., (1999), ^{1}H NMR Spectra of Some Amidrazone Derivatives. **Turk J Chem,** 23, 263 - 267.

Baker, W., (1933), Molecular rearrangement of some o-acyloxyacetophenones and the mechanism of the production of 3-acylchromones. **J. Chem. Soc**., 1381-1389

Baldwin, J. E. (1976), Rules for ring closure. **Journal of the Chemical Society Chemical Communications**, 18, 734-736.

Banholzer, K. and Schmid, H., (1954), Nachweis der intramolekularen Natur der Baker-Venkataraman-Umlagerung , **Helvetica Chimica Acta**, 37, 1706-1716

Baxter, H. and Harborne JB, (1999), **The handbook of natural flavonoids**. Vol.1-2. New York: John Wiley and son.

Billet, D., Lecocq, J., Mentzer, C., Meunier, P.and Xuong, D., (1945), Bull. Soc. **Chim. Fr**., 12, 430.

Belluti, F., Bisi, A., Cavalli, A., Gobbi, S., Hartmann, R. W., Rampa, A., Recanatini, M., Paluszcak, A.and Piazzi, L., (2006), Lead Optimization Providing a Series of Flavone Derivatives as Potent Nonsteroidal Inhibitors of the Cytochrome P450 Aromatase Enzyme. **J. Med. Chem**., 49, 4777-4780.

Bhat, A. S., Whetstone, J. L. and Brueggemeier, R. W. (1999), Novel synthetic routes suitable for constructing benzopyrone combinatorial libraries, **Tetrahedron Lett**., 40, 2469-2472.

Becker, H., Eicher, Th. and Zinsmeister, H. D., (1991), Angew.**Chem. Int. Ed. Engl**., 30, 130.

Beney, C., Boumendjel, A. and Hadjeri, M., (2003), Recent Advances in the Synthesis of Conveniently Substituted Flavones,Quinolones, Chalcones and Aurones: Potential Biologically Active Molecules. **Current Organic Chemistry**, 7, 679-689.

Bhushan, V., Chakrabarti, R., Jajoo, H. K., Lohray, B. B., Madhavan, G. R., Mamidi, R. N., Murali, N., Rajagopalan, R., Rajesh, B. M., Rao, K. N., Rao, P. B., Reddym, A. K., Reddy, P. G., Vikramadithyan, R. K. and Subramaniam, S., (1998), Novel euglycemic and hypolipidemic agents, **J. Med. Chem**., 41, 1619-30.

Biswas, S. K., Kirkham, P. A and Rahman, I., (2006), Regulation of inflammation and redox signaling by dietary polyphenols. **Biochem. Pharmacol**., 30, 1439-52..

Blumberg, J. B. , Graf, B. A. and Milbury, P. E.,(2005), **J. Med. Food**, 8, 281.

Boelens, P. G., Hoorn, D. E. V., Leeuwen, P. A. V.,Nijveldt, R. J., Nood, E. V. and Norren, K. V. (2001), Flavonoids: a review of probable mechanisms of action and potential applications. **American Journal of Clinical Nutrition**, 74, 418-425.

Bonina, F., Castelli, F., Saija, A., Scalese, M., Lanza, M. and Marzullo, D., (1995) Flavonoides As Antioxidant Agents: Importance Of Their Interactio With Biommbranes. **Free Radical Biology and Medicine**, 19, 481-486.

Bourne, G. T., Horton, D. H., and Smythe, M. L., (2003), The Combinatorial Synthesis of Bicyclic Privileged Structures or Privileged Substructures. **Chem. Rev.**, 103, 893-930.

Bracke, M. E., Bisht, K. S., Courtens, A., Jain, R., Jain, S. C., Kumar, A., Kumar, N., Kumar, R., Malhotra, S., Mareel, M. M., Olsen, C. E., Parmar, V. S., Philippe, J., Rajwanshi, V. K,. Sharma, N.K., Sharma, S. K., Singh, S. K., Vennekens, K., van Mark,V. and Wengel, J, (1997), Anti-invasive activity of alkaloids and polyphenolics in vitro. **Bioorg. Med. Chem**., 5, 1609-1619.

Brehm, O., Odenthal, K. P. and Rouwald, H. W. (1994), Planta Med., 60, 101.

Brendel, M.D., Daniel, H., Kuntz, S. and Wenzel, U., (2000), Dietary flavone is a potent apoptosis inducer in human colon carcinoma cells. **Cancer Research**, 60, 3823–3831.

Brinkworth, RI., Fairlie, D.P. and Stoermer, M.J., (1992), Flavones are inhibitors of HIV-1 proteinase. **Biochem Biophys Res Commun**. 188, 7-631.

Broekkamp, C. L. E., Leysen, D., Peeters, B. W. and Pinder, R. M. (1995), Prospects for improved antidepressants. **J. Med. Chem.**, 38, 4615-4633.

Brown, J.P., (1980), A review of the genetic effects of naturally occurring flavonoids, anthraquinones and related compounds. **Mutat Res**., 75, 77-243.

Buchholz, H., Carola, C., Koniga, B., and Walenzyk, T., (2005), Chromone derivatives which bind to human hair. **Tetrahedron**, 61, 7366-7377.

Butler, R. N. and Scott, F. L. (1970), Versatile reactive intermediates: hydrazidic halides. **Chemistry & Industry**, 38, 21-1216.

Carballido- Reboredo, R. Seijas, J. A. and Vazquez-Tato, M. P., (2005**), J. Org. Chem**. , 70, 2855.

Capasso, F,. Di Carlo, G., Izzo, A. A. and Mascolo, N., (1999), Flavonoids: old and new aspects of a class of natural therapeutic drugs**. Life Sci**., 65, 337-353.

Chabot, GG., Dauzonnel, D., FolEasl, B. and Martinez, L., (1997), Synthesis and in vitro cytotoxicity of a series of 3-aminoflavones. **Eur J Med Chem**, 32, 7 l-82.

Chen, Y., Jia Z., Ju, Y. and Zheng, R., (1990), Flavonoids as superoxide scavengers and antioxidants. Free Radic. **Biol. Med.,** 9, 19-21.

Chithan K. and Middleton Jr E, (1993), **The impact of plant flavonoids on mammalian biology: implications for immunity, inflammation and cancer**. In: Harborne JB, editor. The flavonoids: advances in research since 1986. London, UK: Chapman and Hall.

Clemens, F., Drutkowski, G., Frohberg, P. and Wiese, M., (2001), The inactivation of lipoxygenase-1 from soybeans by amidrazones. **Biochimica et Biophysica Acta,** 1549, 88-98.

Cox, J. S. G., (1967), **Nature**, 216, 1328.

Cox, J. S. G., Gwilliam, J., Orr, T. S. C. and Pollard, M. C., (1970), **Celin. Exp. Immunol**., 7, 745.

Cradock, J. C., Grieshaber, C. K., Plowman, J.and Zaharko, D. S., (1986), **Cancer Treatment Reports**, 70, 1415.

Crespo, I., Collado, P. S., Esteller, A., Garcia-Mediavilla, V., Gonzalez-Gallego, J., Sanchez-Campos, S. and Tunon, M. J., (2007), **Eur. J. Pharmacol**., 557, 221.

Cunningham, B. D. M., Dale, I.L., Groundwater, P.W., Hickman, J.A. and Threadgill, M.D., (1992), **Anti Cancer Drug Des .,** 7, 365-384.

Cushman, M., Burg, D, L., Geahlen, R. L. and Nagarathnam, D., (1991), Synthesis and protein-tyrosine kinase inhibitory activities of flavonoid analogues, **J. Med. Chem**., 34, 798-806.

Cushman, M., Geahlen, R. L., Kraker, A. J. and Zhu, H., (1994), **J. Med. Chem**., 37, 3353-3362.

Cushman, M. and Nagarathan, D., (1991), **Tetrahedron**, 47, 5071.

Cushnie, T. P. T. and Lamb, A. J., (2005), Antimicrobial activity of flavonoids. **International Journal of Antimicrobial Agents**, 26, 343–356.

Daniel, H., Kuntz, S. and Wenzel, U., (1999), Comparative analysis of the effects of flavonoids on proliferation, cytotoxicity, and apoptosis in human colon cancer cell lines. **Eur J Nutr**, 38, 133–142.

Dahlen, K., Grotli, M. and Luthman, K., (2006),**Synlett**, 897.

De Groot, H., (1994), Reactive oxygen species in tissue injur. **Hepatogastroente - rology**, 41, 328-32.

Demuynck, L., Fabis, F., Fossey, C., Lecoutey, C., Lefoulon, F. and Rault, S., (2008), A convenient microwave-assisted 5-amination of flavones,**Tetrahedron**, 64, 11243-11248.

Dixon, S., Lam, K. S., Song, A., Wang, X. and Yao, N., (2007), **J. Comb. Chem**., 9, 668.

Dow, R. L. and Kreutter, D. K. (1995), **Ann. Rep. Med. Chem**., 30, 159.

Dubrovsky, A. V. and Larock, R. C., Zhou, C., (2006), **J. Org. Chem.**, 71, 1626;

Ducrot, P. H., Es-Safi, N. and Ghidouche., S., (2007), Flavonoids: Hemisynthesis, Reactivity, Characterization and Free Radical Scavenging Activity. **Molecules**, 12, 2228-2258.

Edwards, A. M. and Howell, J. B. L., (2000), The chromones: history, chemistry and clinical development. A tribute to the work of Dr R. E. C. Altounyan. **Clinical and Experimental Allergy**, 30, 756-774.

Eicher, T. and Hauptmann, S., (2003), **The Chemistry of Heterocycles**. 2, (261-265), Germany, Wiley-VCH Veriag GmbH & Co. KGaA.

Eicher,Th. and Laas, H. J., (1989), **Journ. Hattori Bot. Lab**., 67, 383.

El-Abadelah, M. M., Hussein, A. Q. and Thaher, B. A. (1991), Heterocycles from nitrile imines. Part IV. Chiral 4,5-dihydro-1,2,4-triazin-6-ones. **Heterocycles**, 32, 1879-1895.

Ellis, G. P., (1977), **In The Chemistry of Heterocyclic Compounds**, 31, (749): Wiley: New York.

Frasinyuk, M. S. and Khilya, V. P., (1999), Preperation and reactions of isoflavone heteroanalogs. **Chemistry of Heterocyclic Compounds**, 35, 3-23.

Gilchrist, T. L., (1997), **Heterocyclic Chemestry**, (3rd ed.). Harlow, Essex, England: Addison Wesley Longman limited.

Gomi, K., Kasai, M., Morimoto, M., Sagaya, T. and Shida, Y., (1990), **Eur Patent Appl EP 374 789; Chem Abstr .,** 23, 1085.

Grace, PA., (1994), Ischaemia-reperfusion injury., **Br. J. Surg**., 81, 47-637.

Gryglewski, RJ. and Robak, J., (1996), Bioactivity of flavonoids. **Pol. J. Pharmacol**. , 48, 64-555.

Gryglewski, R.J. and Robak, J., (1988), Flavonoids are scavengers of super oxide anions. **Biochem. Pharmacol**. 37, 837-841.

Gupta, S. C. and Mukerjee, S. M., (1973), **Tetrahedron Lett**., 51, 5073.

Harborne, J. B., and Williams, C. A., (2000), Advances in flavonoid research since 1992. **Phytochemistry**, 55, 481-504.

Harley, J. P. and Klein, D. A., (1999), **Prescott LM, Microbiology**. London, UK: WCB/McGraw-Hill.

Havsteen B., (1983), Flavonoids, a class of natural products of high pharmacological potency. **Biochem Pharmacol.**, 32, 1141-1148.

Heusgen, R. (1968), Mechanism of 1,3-dipolar cycloadditions. Reply**. Journal of Organic Chemistry**, 33, 2291-2297.

Higuchi, T., Ikeshiro, Y., Kashiwada, Y., Nakano, T., Sekiya, M., Shibata, H., Takaishi, Y., and Tanaka, N., (2009), Chromone and chromanone glucosides from Hypericum sikokumontanum and their anti-Helicobacter pylori activities. **Phytochemistry**, 70, 141-146.

Hodek , P., Stiborova, M. and Trefil, P., (2002) ,Flavonoids-potent and versatile biologically active compounds interacting with cytochromes P450. **Chemico-Biological Interactions,** 139, 1-21.

Hoult, J. R. S., Moroney, M. A. and Paya, M., (1994), Actions of flavonoids and coumarins on lipoxygenase and cyclooxygenase. **Methods Enzymol**., 234, 443.

Hussein, A. Q., El-Abadelah, M. M. and Sabri, W. S. (1984), Heterocycles from nitrile oxides. II. 1,2,4-Oxadiazin-6-ones. **Journal of Heterocyclic Chemistry**, 21, 455- 459.

Ibarra, M., Hong, E. and Villalobos-Molina, R. (2000), The α-adrenoceptor antagonist, zolertine, inhibits α-1D- and α-1A adrenoceptor-mediated vasoconstriction in vitro. **Journal of Autonomic Pharmacology**, 20, 139-145.

Ishar, M. P. S., Singh, G., Singh, L., (2002), 2-(N-Methylanilino)-3-formylchromone-a versatile synthon for incorporation of chromone moiety in a variety of heterocyclic systems and macrocycles through reactions with bifunctional nucleophiles, **Tetrahedron**, 58, 7883 -7890**.**

Ishchenko,V. V. and Khilya, V. P., (2002), Flavones, Isoflavones, And 2- and 3-hetarylchromones In Reactions With Hydroxylamine(review). **Chemistry of Heterocyclic Compounds**, 38, 883-897.

Iversen, L., Kim, HP., Mani, I. and Ziboh, VA.,(1998), Effects of naturally-occurring flavonoids and bioflavonoids on epidermal cyclooxygenase and lipoxygenase from guinea-pigs. **Prostaglandins Leukot Essent Fatty Acids**. 58, 17 24.

Jiang, X., Song, Y. and Long, Y. (2004), Facile synthesis of 4-substituted-4-aminopiperidine derivatives, the key building block of piperazine-based CCR5 antagonists. **Bioorganic & Medicinal Chemistry Letters**, 14, 3675-3678.

Joule, J. A. and Mills, K., (2000), **Heterocyclic Chemistry**, (4th ed.). Oxford; Malden, MA: Blackwell science Ltd.

Jovanovic, S. V.; Marjanovic, B.; Simic, M. G., Steenken, S. and Tosic, M., (1994), **J. Am. Chem. Soc.**, 116, 4846.

Kabalka, G. W. and Mereddy, A. R., (2005), Microwave Assisted Synthesis of Functionalized Flavones and Chromones **Tetrahedron Lett**, 46, 6315-6317.

Kasum, C. M. and Ross, J. A., (2002), Dietary flavonoids: bioavailability, metabolic effects, and safety. **Annu. Rev. Nutr**, 22, 19-34.

Katritzky, A. R., Nie, P. L., Dondoni, A. and Tassi, D. (1979), Heterocycles in organic synthesis. Part 24. A new synthesis of NN'-diarylcarbodi-imides. **Journal of the Chemical Society, Perkin Transactions 1: Organic and Bio-Organic Chemistry**,

8, 1961-1963.

Khalaj, A., Adibpour, N., Shahverdi, A. R. and Daneshtalab, M. (2004), Synthesis and antibacterial activity of 2-(4-substituted phenyl)-3(2H)-isothiazolones. **Eur. J. Med. Chem.**, 39, 699-705.

Kosmider, B. and Osiecka, R., (2004) Flavonoid Compounds: A Review of Anticancer Properties and Interactions with cis-Diamminedichloroplatinum(II). **Drug Development Research**, 63, 200–211.

Kumar, K. H. and Perumal, P. T., (2007), A novel one-pot oxidative cyclization of 2′-amino and 2′-hydroxychalcones employing FeCl3·6H2O–methanol. Synthesis of 4-alkoxy-2-aryl-quinolines and flavones. **Tetrahedron**, 63, 9531 -9535.

Kumar, R. and Yusuf, M., (2006), Chromones and bischromones: an account of photoinduce dreactions. **ARKIVOC**, 9, 239-264.

Lee, S.F. and Xu, H.-X., (2001), Activity of plant flavonoids against antibiotic-resistant bacteria, **Phytother. Res.**, 15, 39–43.

Machado, N.F.L. and Marques, M.P.M., (2010), Bioactive Chromone Derivatives - Structural Diversity. **Current Bioactive Compounds**, 6, 76-89.

Martens, S. and Mithofer, A., (2005), Flavones and flavone synthases. **Phytochemistry**, 66, 2399-2407.

Marder, M., Viola, H., Bacigaluppo, J. A., Colombo, M. I., Wasowski, C., Wolfman, C., Medina, J. H., Ruveda, E. A. and Paladini, A.C., (1998), **Biochem. Biophys. Res. Commun**., 249, 481.

Miao, H. and Yang, Z. (2000), Regio-specific carbonylative annulation of iodophenol acetates and acetylenes to construct the flavones by a new catalyst of palladiu-thiourea-dppp complex. **Org. Lett.,** 2, 1765.

Mubarak, M. S. and Ayoub, M. T. (2007), Recent Approaches In The Synthesis And Reactions Of Pyranones And Their Benzoderivatives. **Modern Approaches to the Synthesis of O- and N-Heterocycles,** vol 1, 47-97. Teodor S. K. and Enrique L. L. eds., Research Signpost: Trivandrum, India.

Mustapha I. C., (1879), **R Acad Sci Paris**; 89, 442.

Naito, H., Ohsuki, S., Atsumi, R., Minami, M., Mochizuki, M., Hirotani, K., Kumazawa, E. and Ejima, A. (2005), Synthesis and antitumor activity of novel pyrimidinyl pyrazole derivatives. III. Synthesis and antitumor activity of 3- phenylpiperazinyl-1-trans-propenes. **Chemical & Pharmaceutical Bulletin**, 53, 153-163.

Phillips, R. R.. (1959), The Japp-Klingeman reaction. **Organic Reactions**, 10, 143-178.

Raw, S. A. and Taylor, R. J. K. (2010), Recent advances in the chemistry of 1,2,4-triazines. **Advances in Heterocyclic Chemistry**, 100, 75-100.

Sakarkar, D. M., Kakde, R. B. and Tapas, A. R., (2008), Flavonoids as Nutraceuticals. **Tropical Journal of Pharmaceutical Research**, 7, 1089-1099.

Selway, J. W. T., (1986), Antiviral activity of flavones and flavans. In: Cody V, Middleton E, Harborne JB, editors. **Plant flavonoids in biology and medicine**: **biochemical, pharmacological, and structure–activity relationships**. New York, NY: Alan R. Liss, Inc.

Sharp, D. B. and Hamilton, C. S. (1946), Derivatives of 1,2,4-triazole and of pyrazole. **Journal of the American Chemical Society**, 68, 588-591.

Shawali, A. S. (1993), Reactions of heterocyclic compounds with nitrilimines and their precursors. **Chem. Rev.**, 93, 2731–2777.

Shawali, A. S. and Abdelhamid, A. O. (1976), New routes to aroylthiadiazolines and arylazothiazoles from phenylglyoxalyl bromide arylhydrazones and phenacyl thiocyanate. **Journal of Heterocyclic Chemistry**, 13, 45-49.

Shawali, A. S. and Parkanyi, C. (1980), Hydrazidoyl halides in the synthesis of heterocycles. **Journal of Heterocyclic Chemistry**, 17, 54-833.

Sosnovskikh, Ya., (2003), Synthesis and reactions of halogen-containing chromones. **V. Russ. Chem. Rev**., 72, 489-516.

Tan, RX., Liu, ZL., Yang, L., Zheng, WF., (1996), Two flavones from Artemisia giraldii and their antimicrobial activity. **Planta Med**., 62,2-160.

Vu, C. B., Peng, B., Kumaravel, G., Smits, G., Jin, X., Phadke, D., Engber, T., Huang, C., Reilly, J., Tam, S., Grant, D., Hetu, G., Chen, L., Zhang, J. and Petter, R. C. (2004), Piperazine Derivatives of [1,2,4]Triazolo[1,5-a][1,3,5]triazine as Potent and Selective Adenosine A2a Receptor Antagonists. **Journal of Medicinal Chemistry**, 47, 4291-4299.

Wawzonek, S., (1950), Chromones, Flavones , And Isoflavones. In: Elderfield, R. C (Ed.), **Heterocyclic Compounds**, (pp.229-235). New York: John Wiley and Sons.

Widmann, O. (1893), New triazole and triazines derivatives. **Berichte der Deutschen Chemischen Gesellschaft**, 26, 2612-2617.

Wolkoff, P., Hammerum, S., Callaghan, P. D. and Gibson, M. S. (1974), Routes to Naryl-N'-thioaroylhydrazines and related sym- and unsym-hydrazonyl sulfides and a note on the so-called N-phenyl-N'-thiobenzoyldiimide. **Canadian Journal of Chemistry**, 52, 879-883.

Yao, H-C. and Resnick, P. (1962), Azo-hydrazone conversion. I. The Japp-Klingemann reaction. **Journal of the American Chemical Society**, 84, 3514-3517.

Yoon, J., Yoo, E. A., Kim, Ji-Y., Pae, A. N., Rhim, H., Park, W.-K., Kong, J. Y. and Park C. H.-Y. (2008), Preparation of piperazine derivatives as 5-HT7 receptor antagonists. **Bioorganic and Medicinal Chemistry** 16, 5405–5412.

7. Appendix : NMR spectra

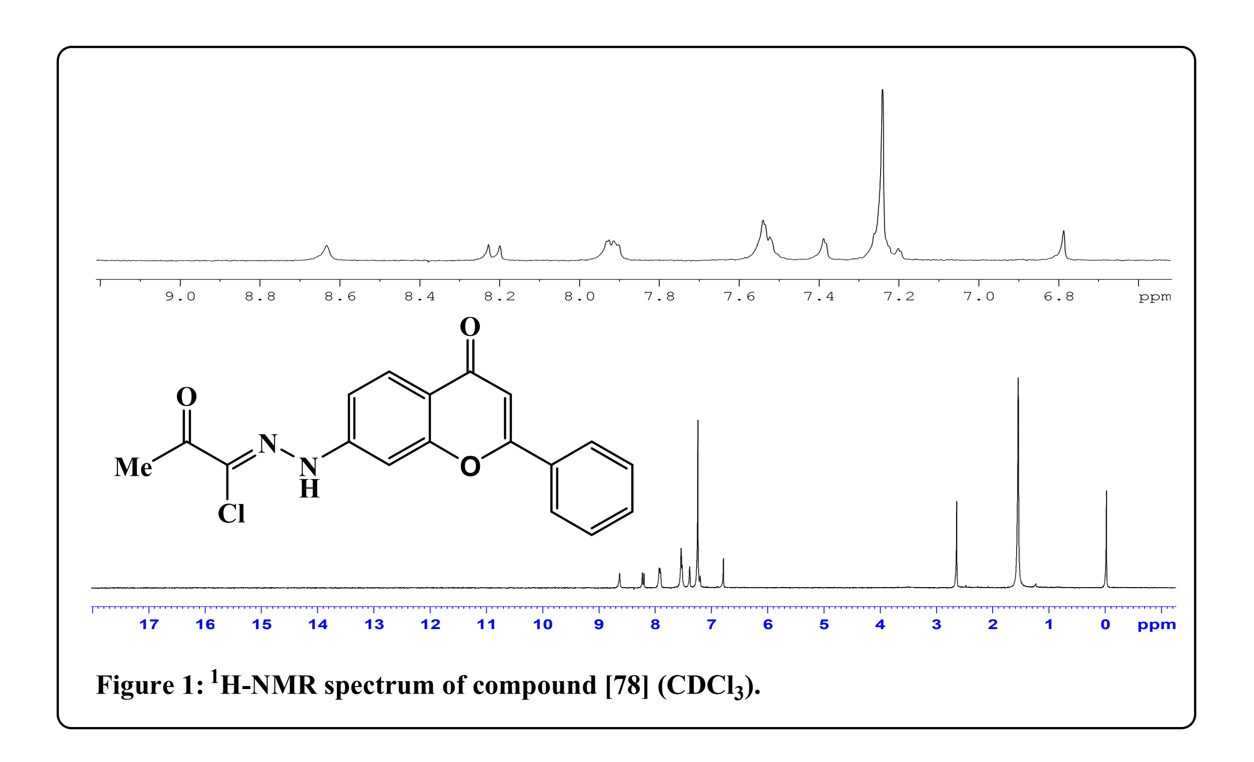


Figure 1: ^{1}H-NMR spectrum of compound [78] ($CDCl_3$).

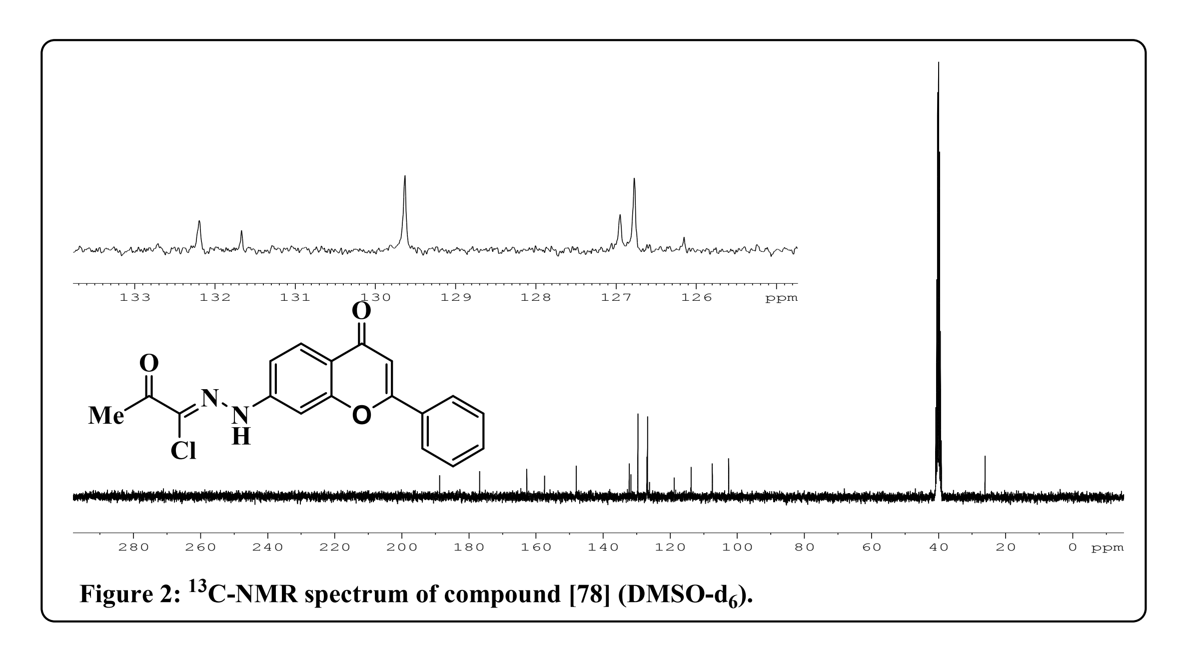


Figure 2: ^{13}C-NMR spectrum of compound [78] (DMSO-d_6).

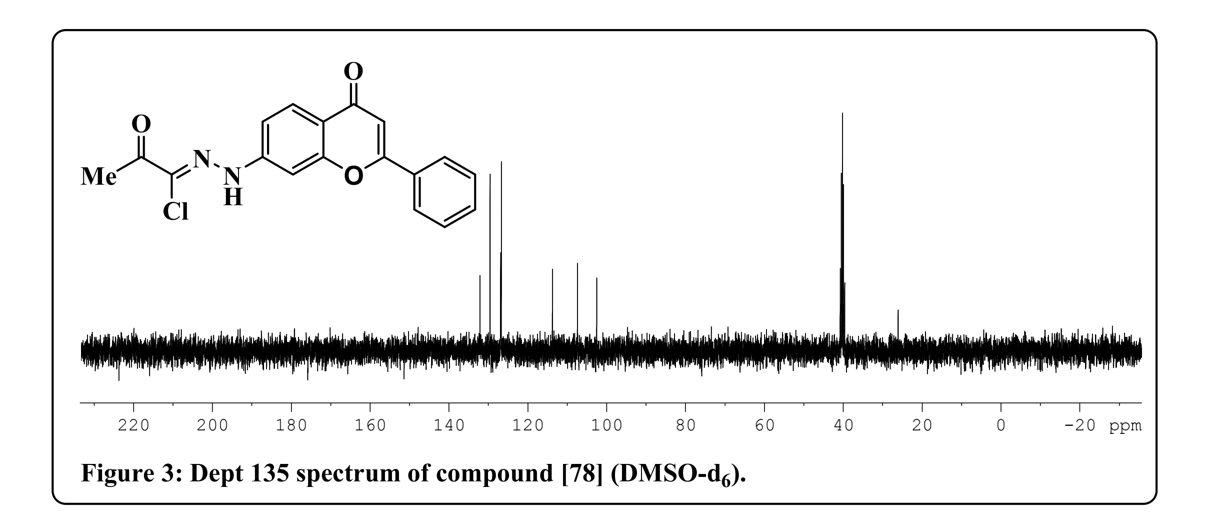


Figure 3: Dept 135 spectrum of compound [78] (DMSO-d$_6$).

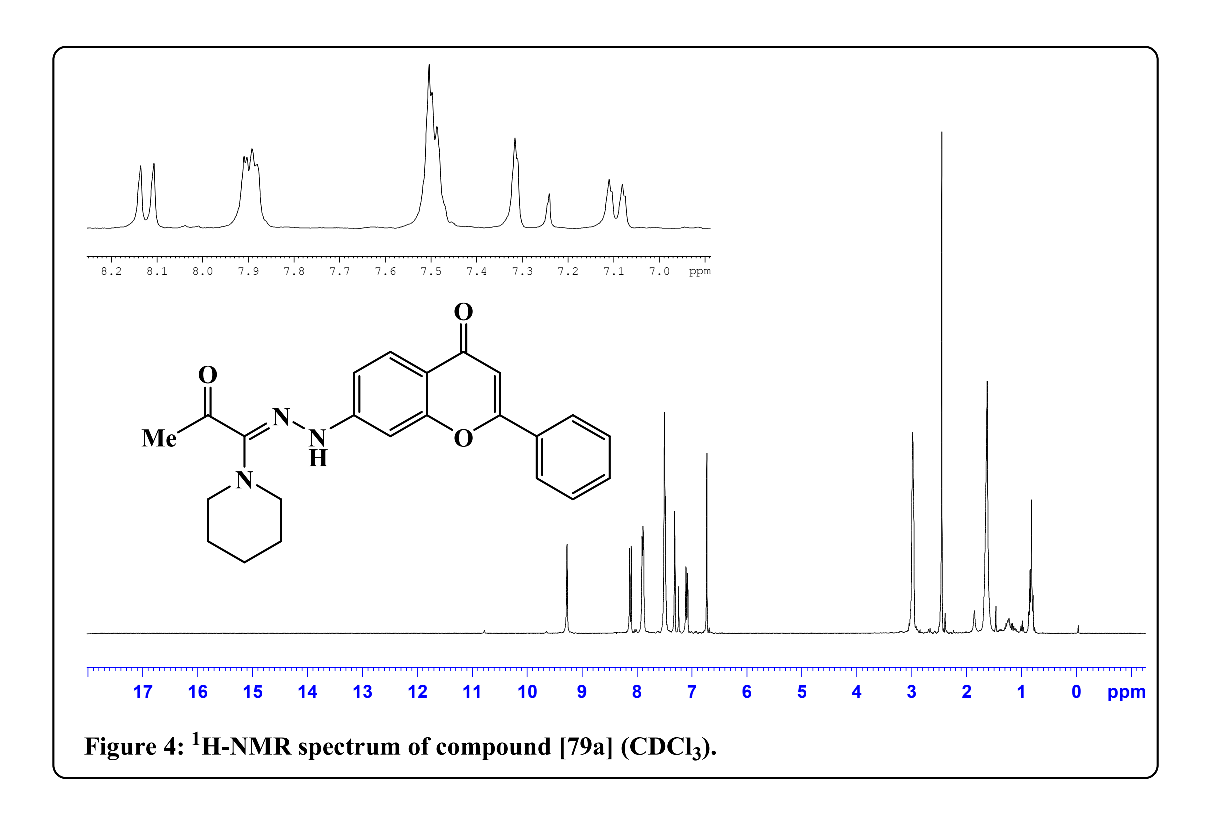


Figure 4: ^{1}H-NMR spectrum of compound [79a] (CDCl$_3$).

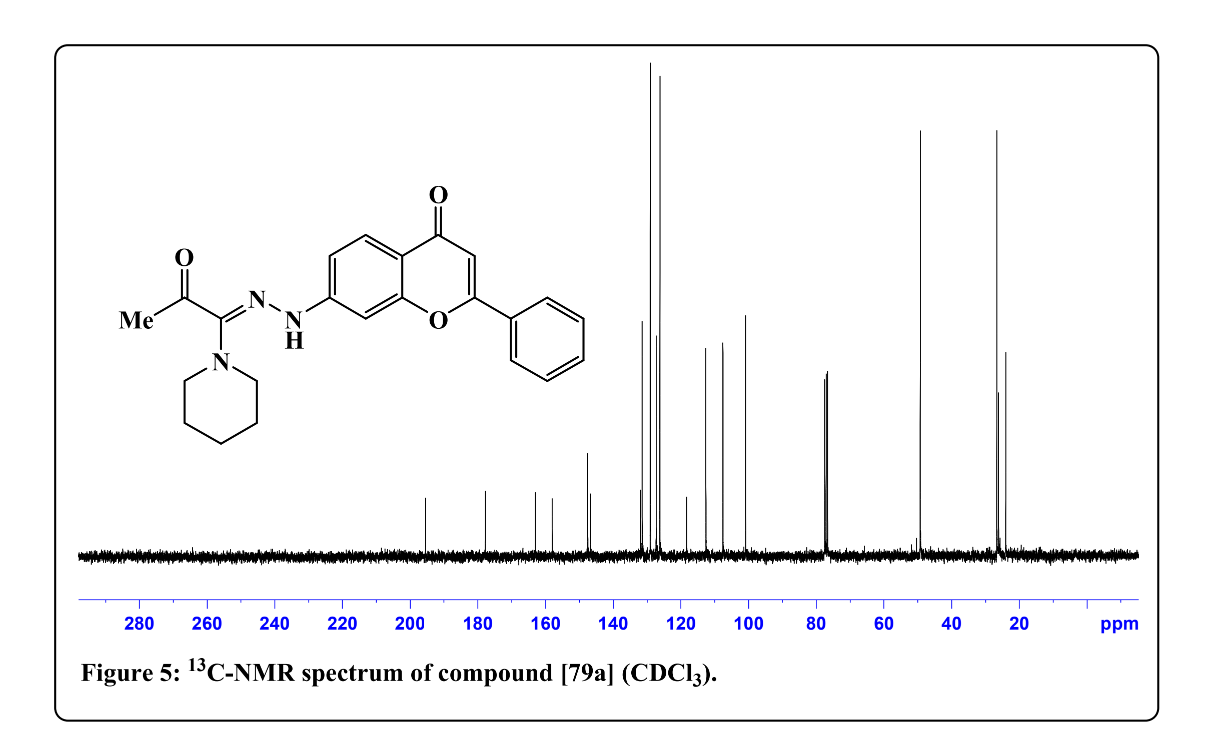


Figure 5: ^{13}C-NMR spectrum of compound [79a] ($CDCl_3$).

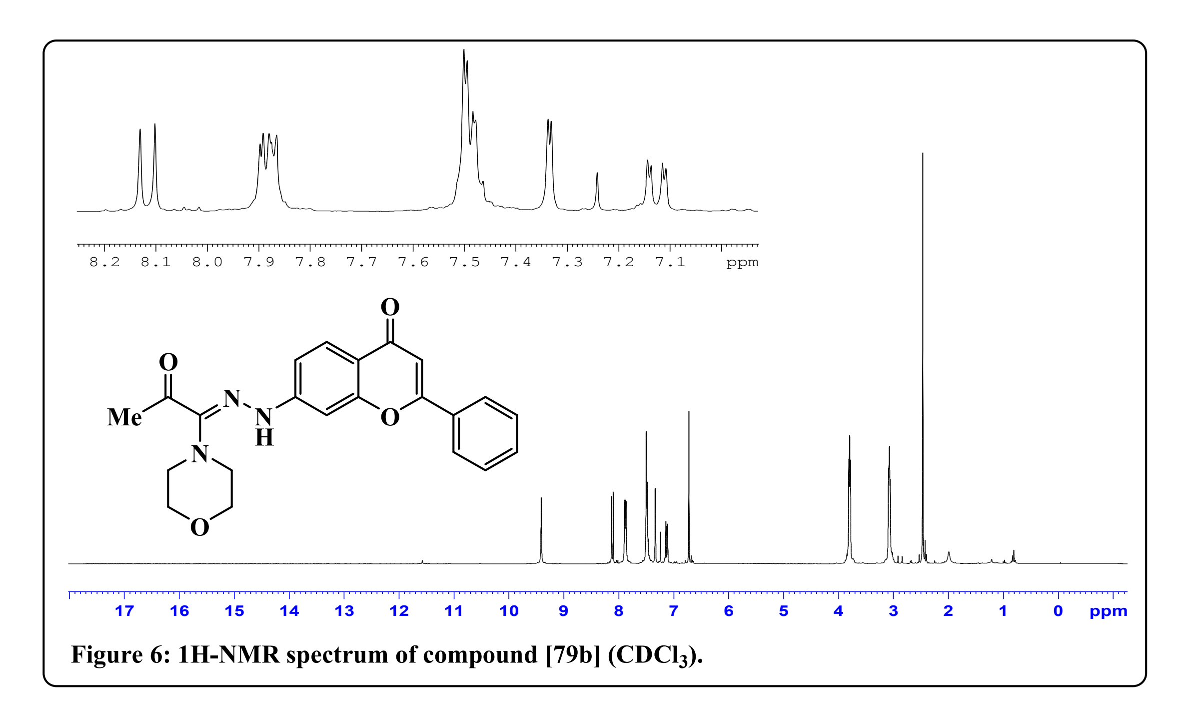


Figure 6: 1H-NMR spectrum of compound [79b] ($CDCl_3$).

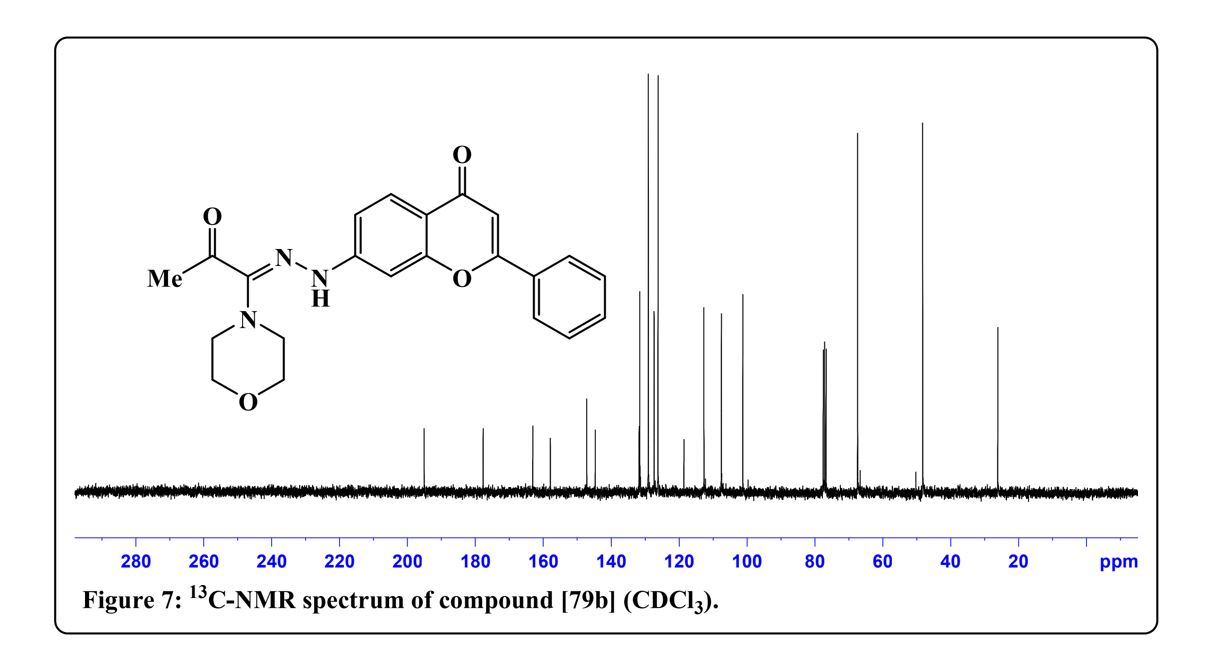


Figure 7: ^{13}C-NMR spectrum of compound [79b] ($CDCl_3$).

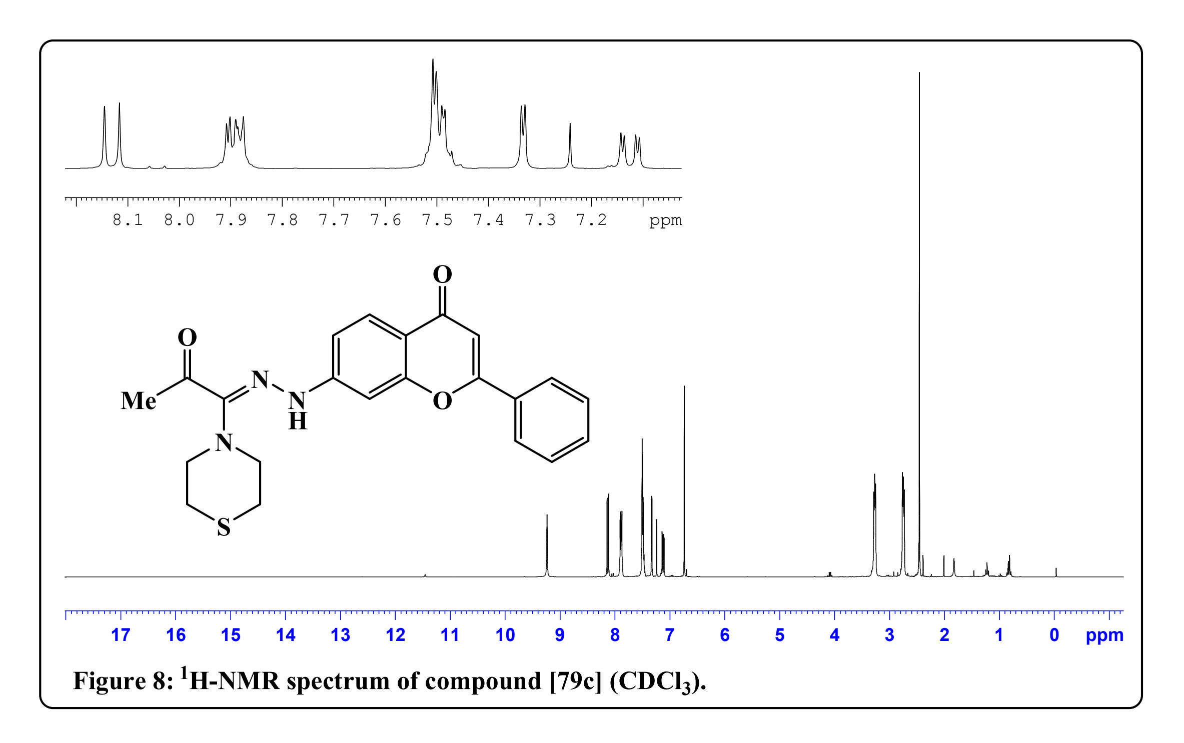


Figure 8: ^{1}H-NMR spectrum of compound [79c] ($CDCl_3$).

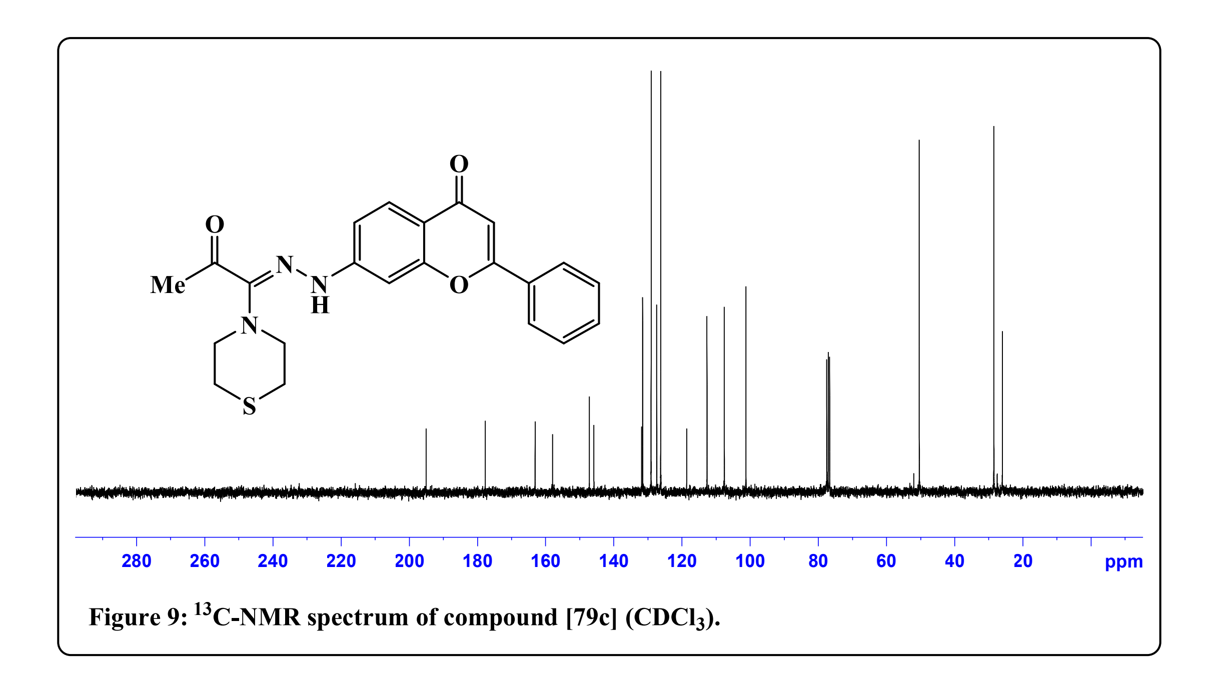


Figure 9: ^{13}C-NMR spectrum of compound [79c] ($CDCl_3$).

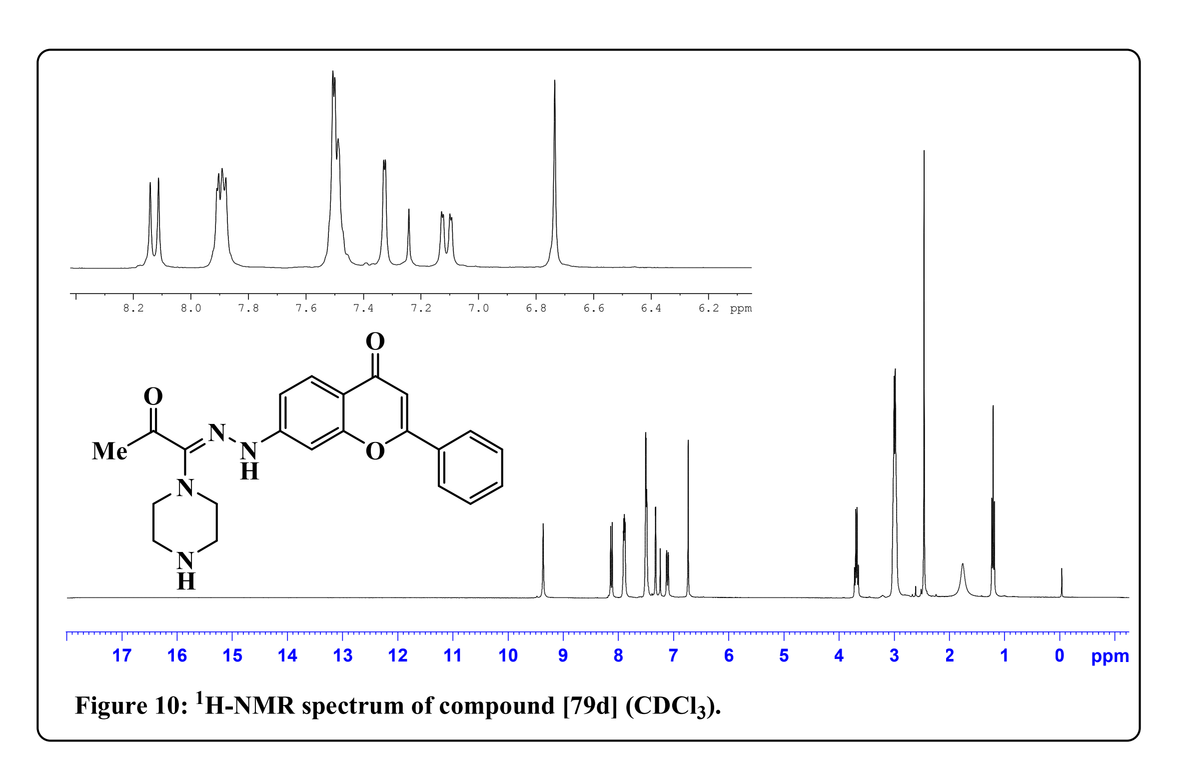


Figure 10: ^{1}H-NMR spectrum of compound [79d] ($CDCl_3$).

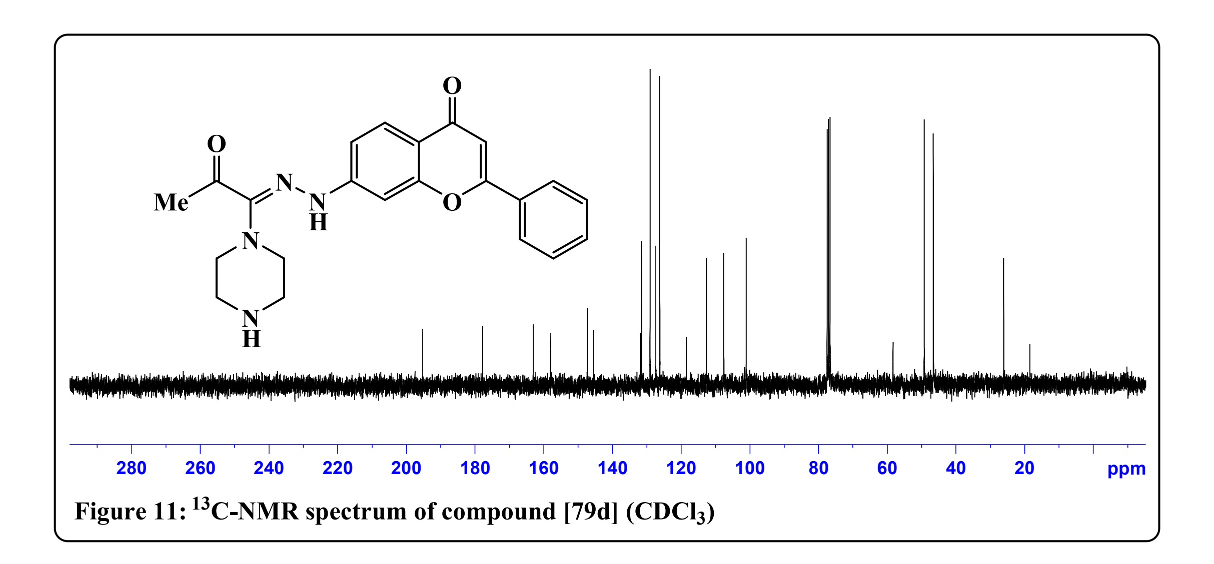


Figure 11: ^{13}C-NMR spectrum of compound [79d] ($CDCl_3$)

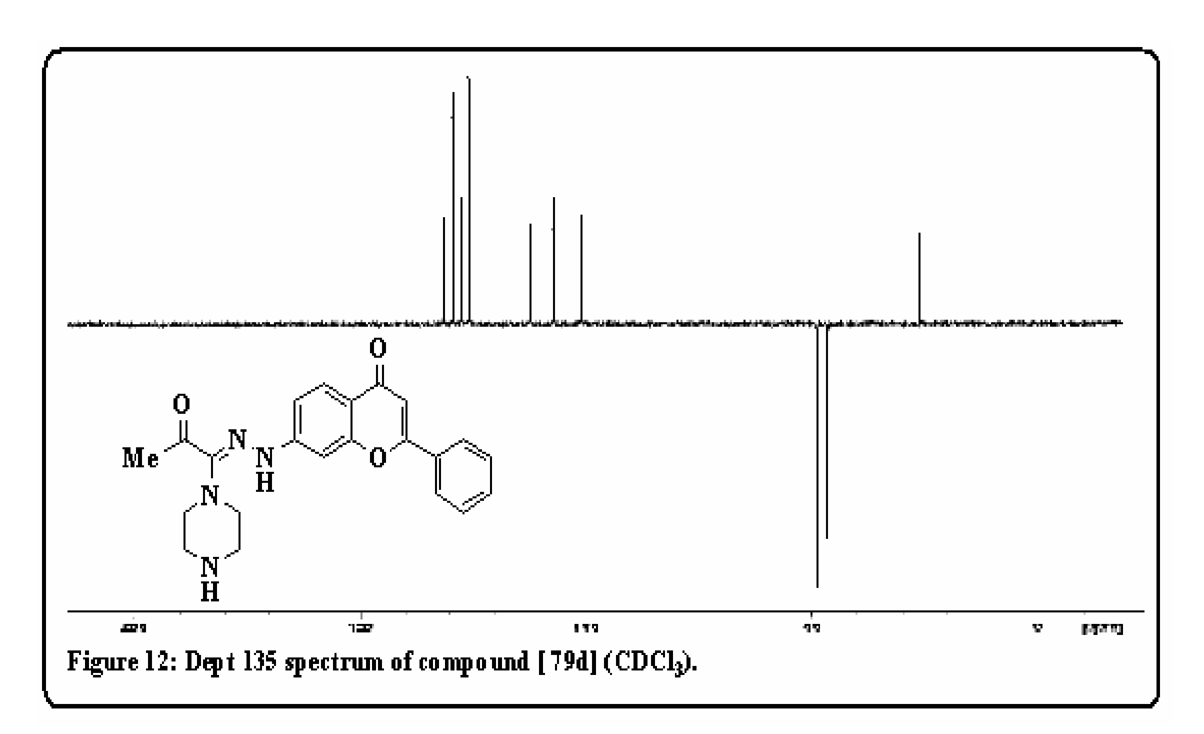


Figure 12: Dept 135 spectrum of compound [79d] ($CDCl_3$).

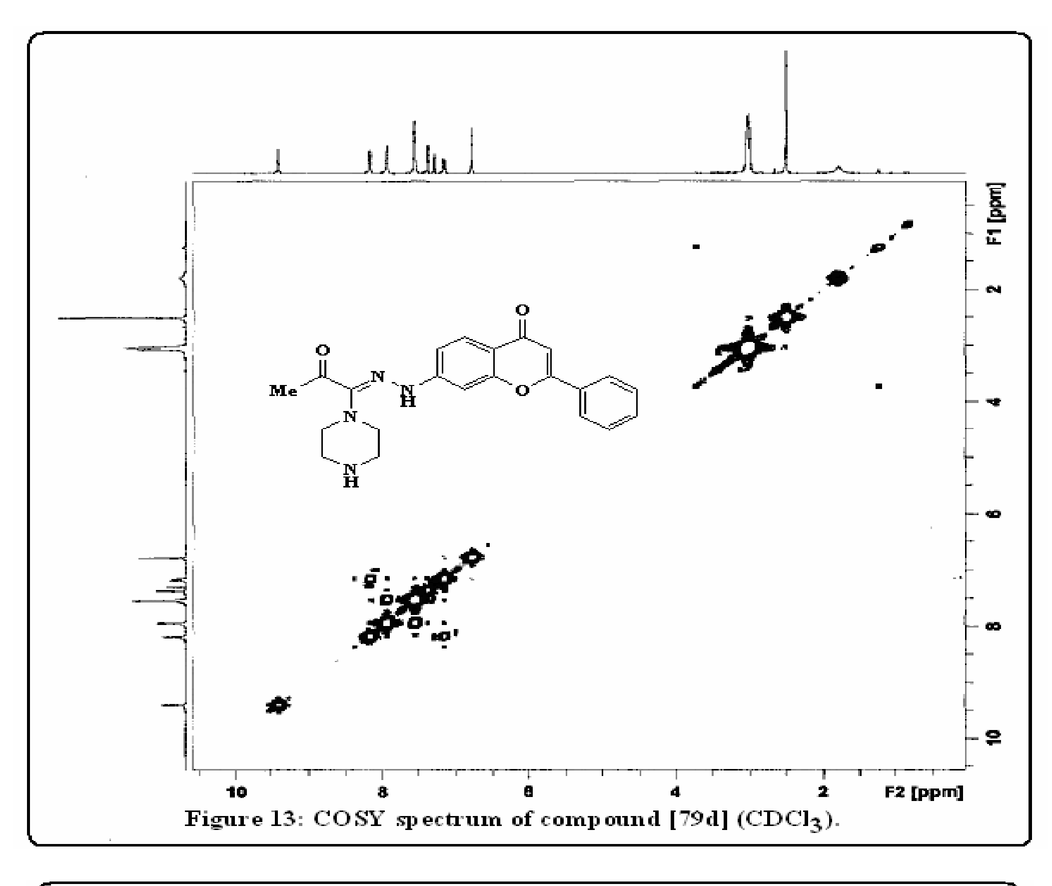


Figure 13: COSY spectrum of compound [79d] ($CDCl_3$).

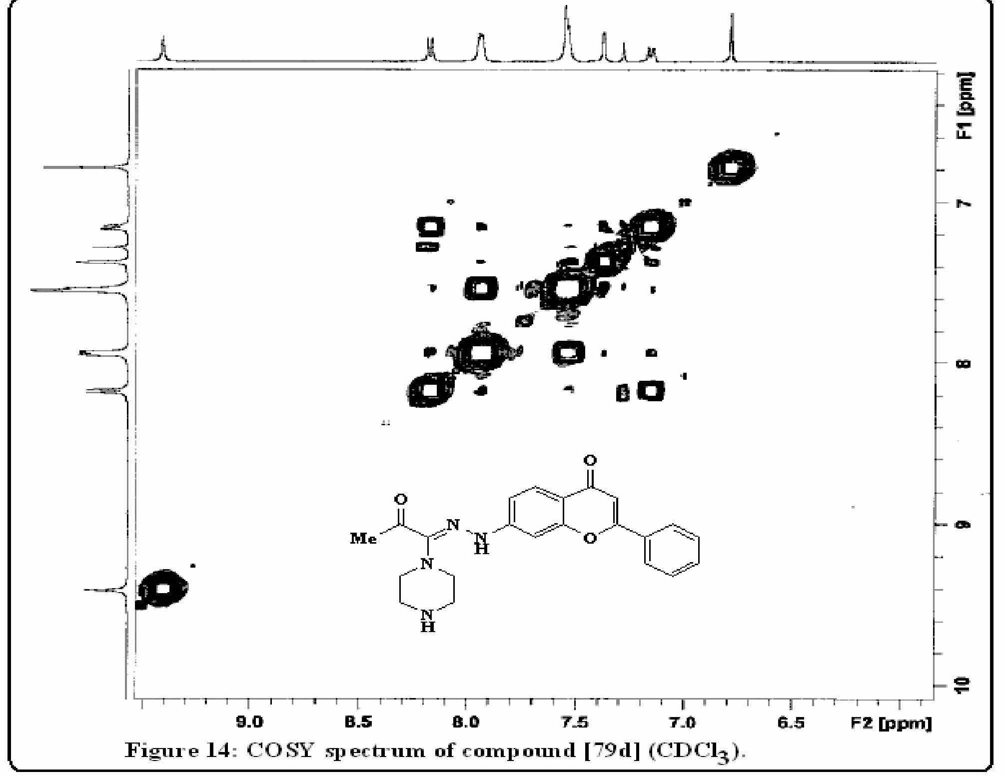


Figure 14: COSY spectrum of compound [79d] ($CDCl_3$).

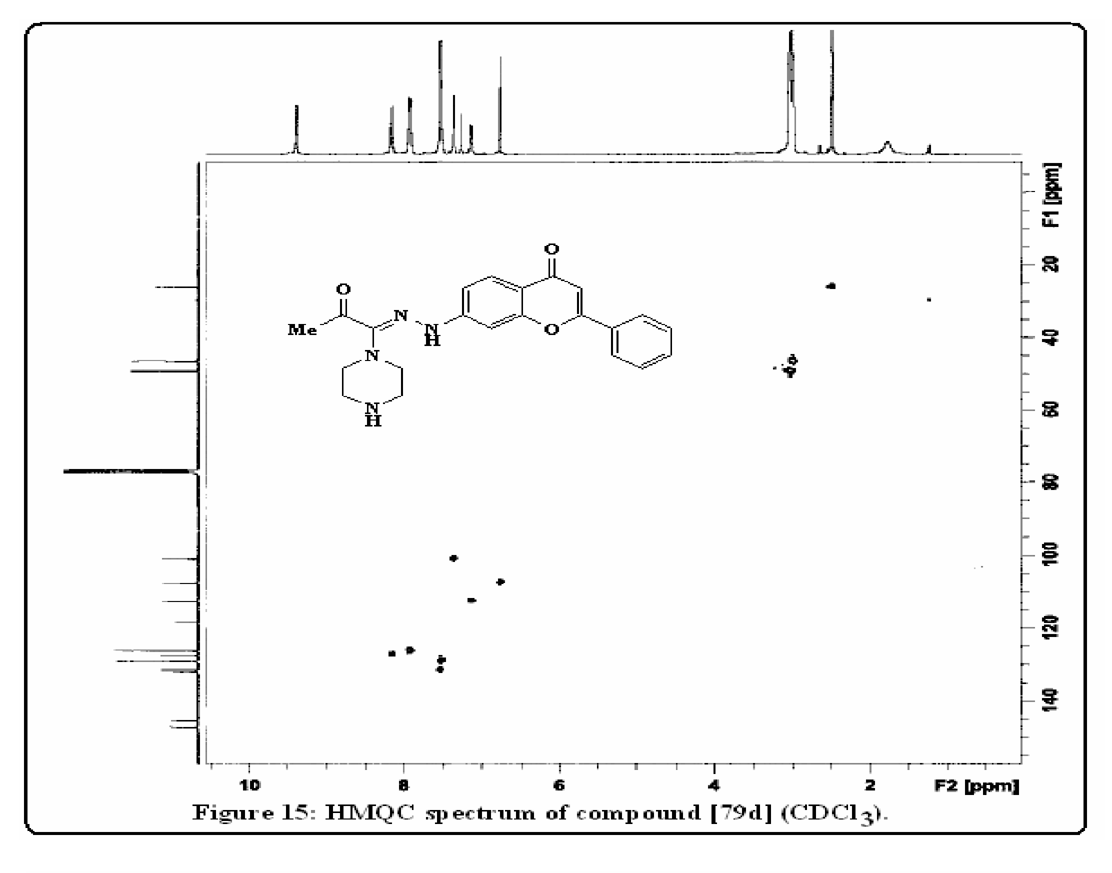


Figure 15: HMQC spectrum of compound [79d] ($CDCl_3$).

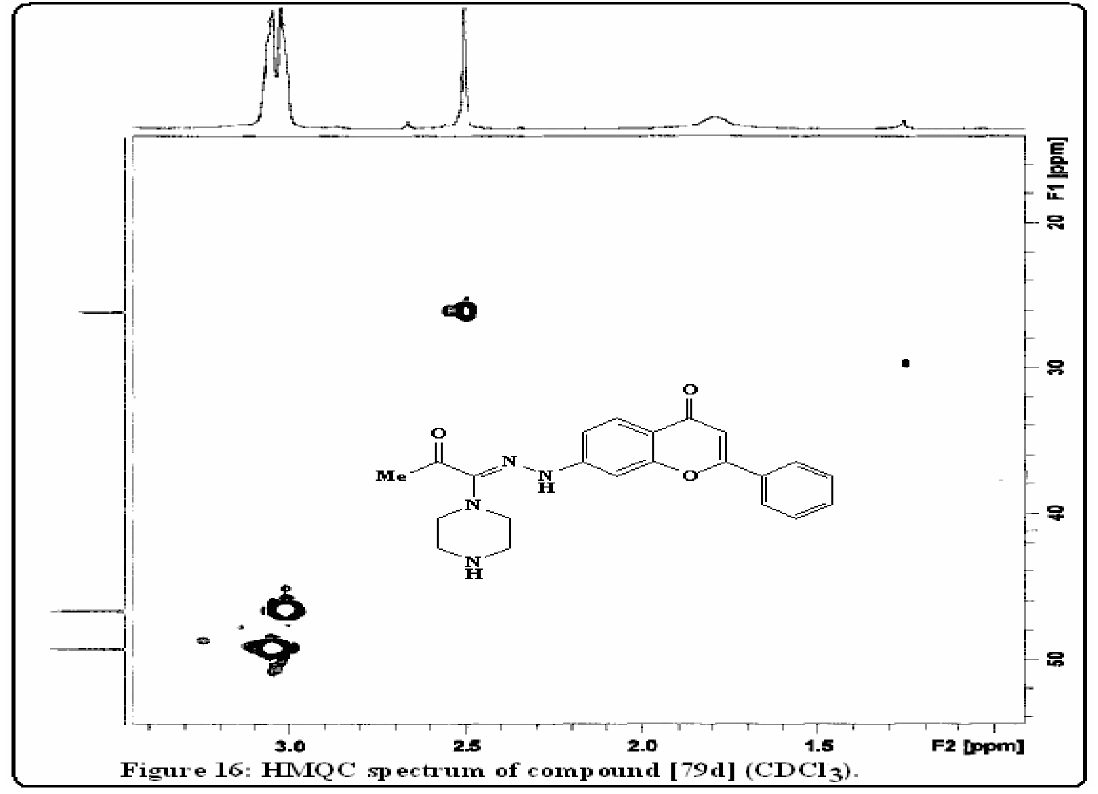


Figure 16: HMQC spectrum of compound [79d] ($CDCl_3$).

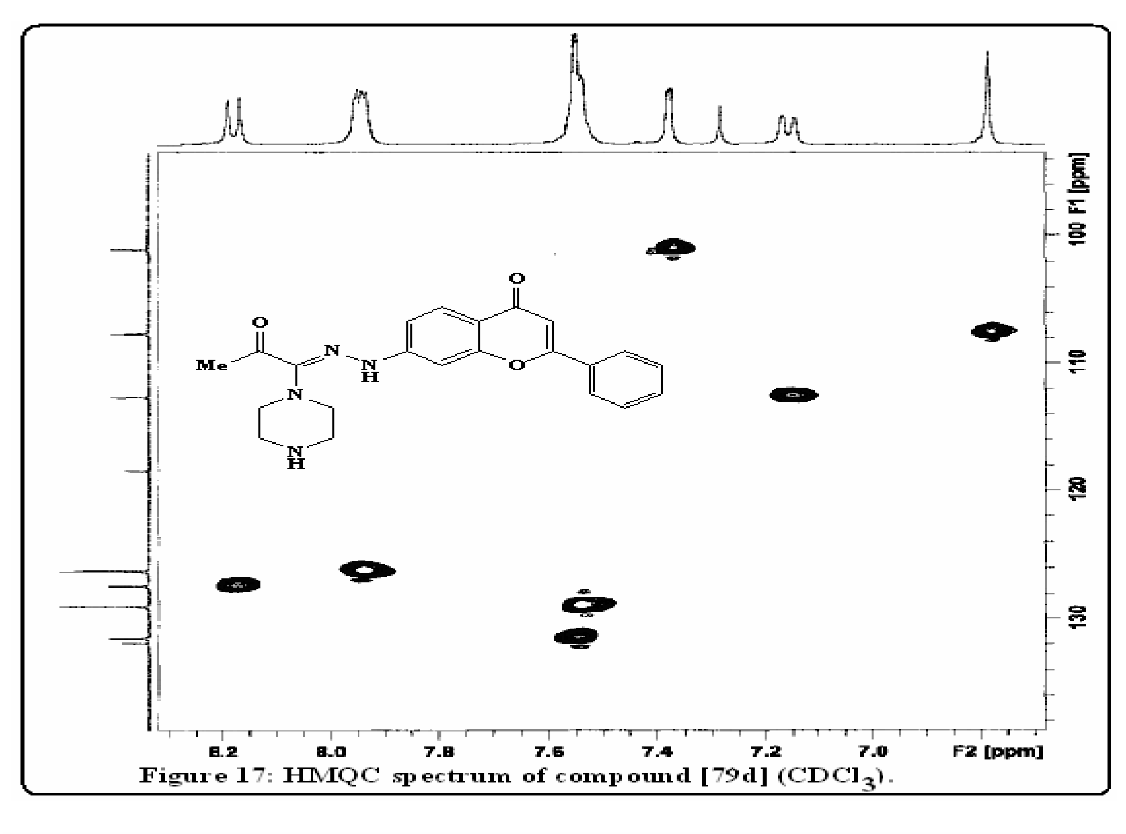


Figure 17: HMQC spectrum of compound [79d] ($CDCl_3$).

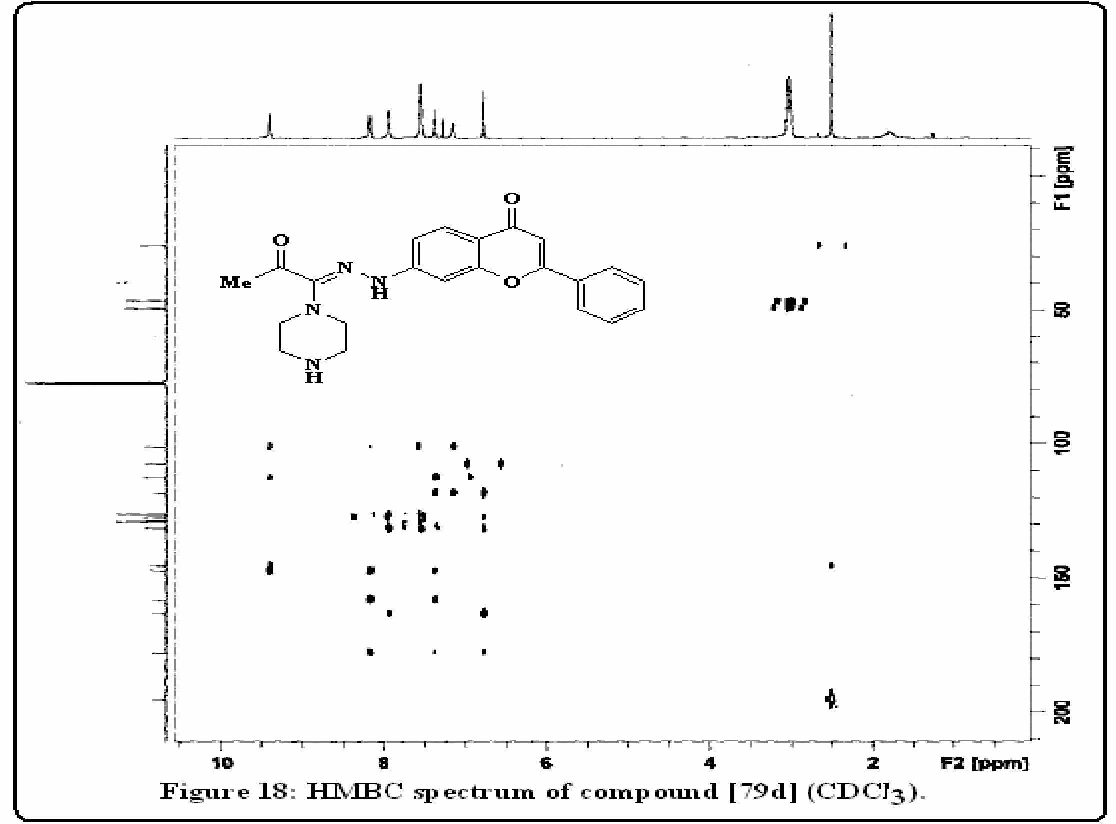


Figure 18: HMBC spectrum of compound [79d] ($CDCl_3$).

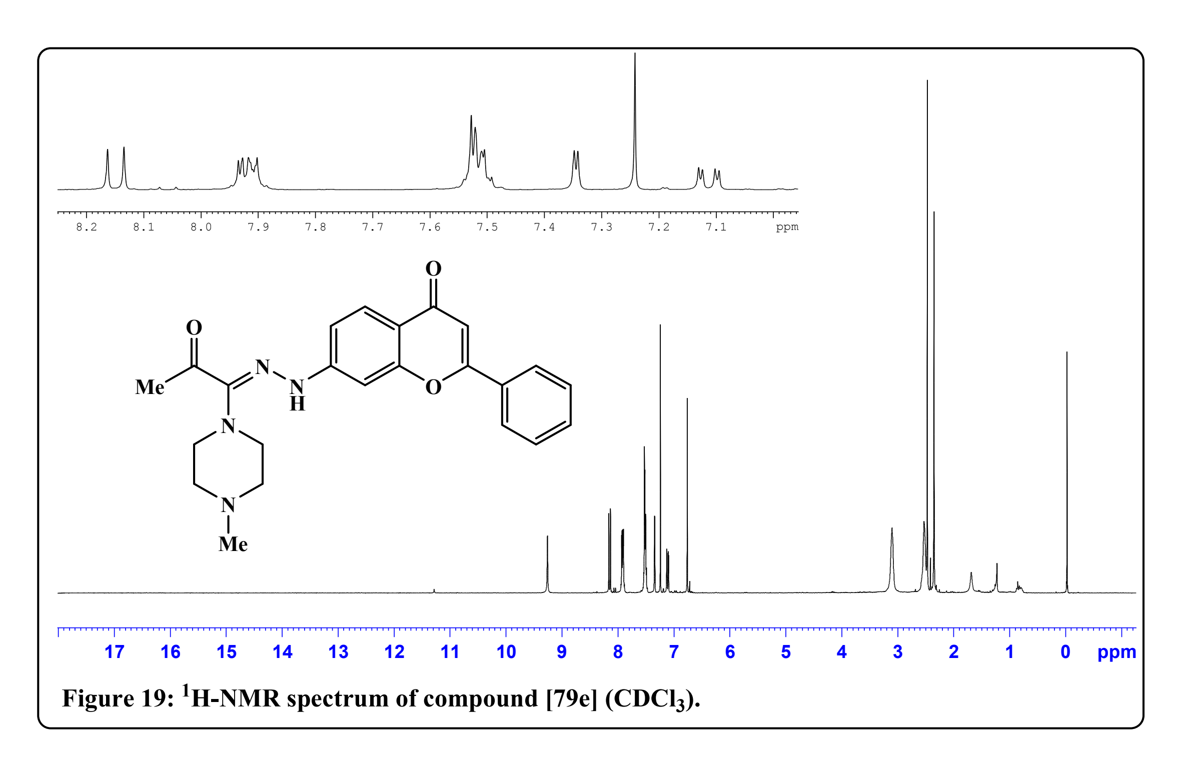


Figure 19: ^{1}H-NMR spectrum of compound [79e] ($CDCl_3$).

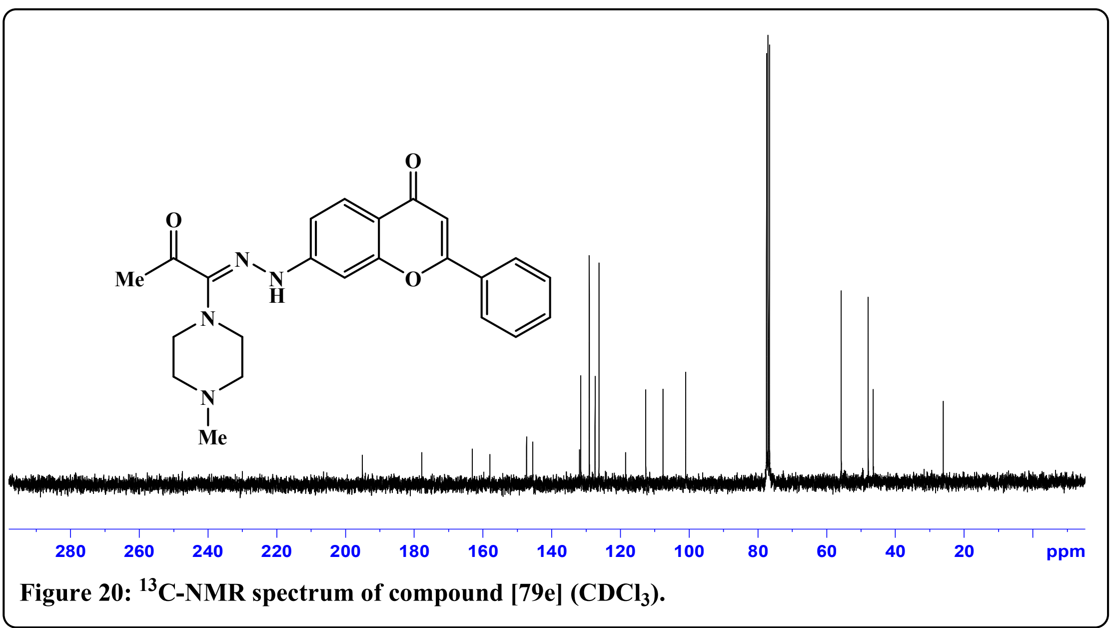


Figure 20: ^{13}C-NMR spectrum of compound [79e] ($CDCl_3$).

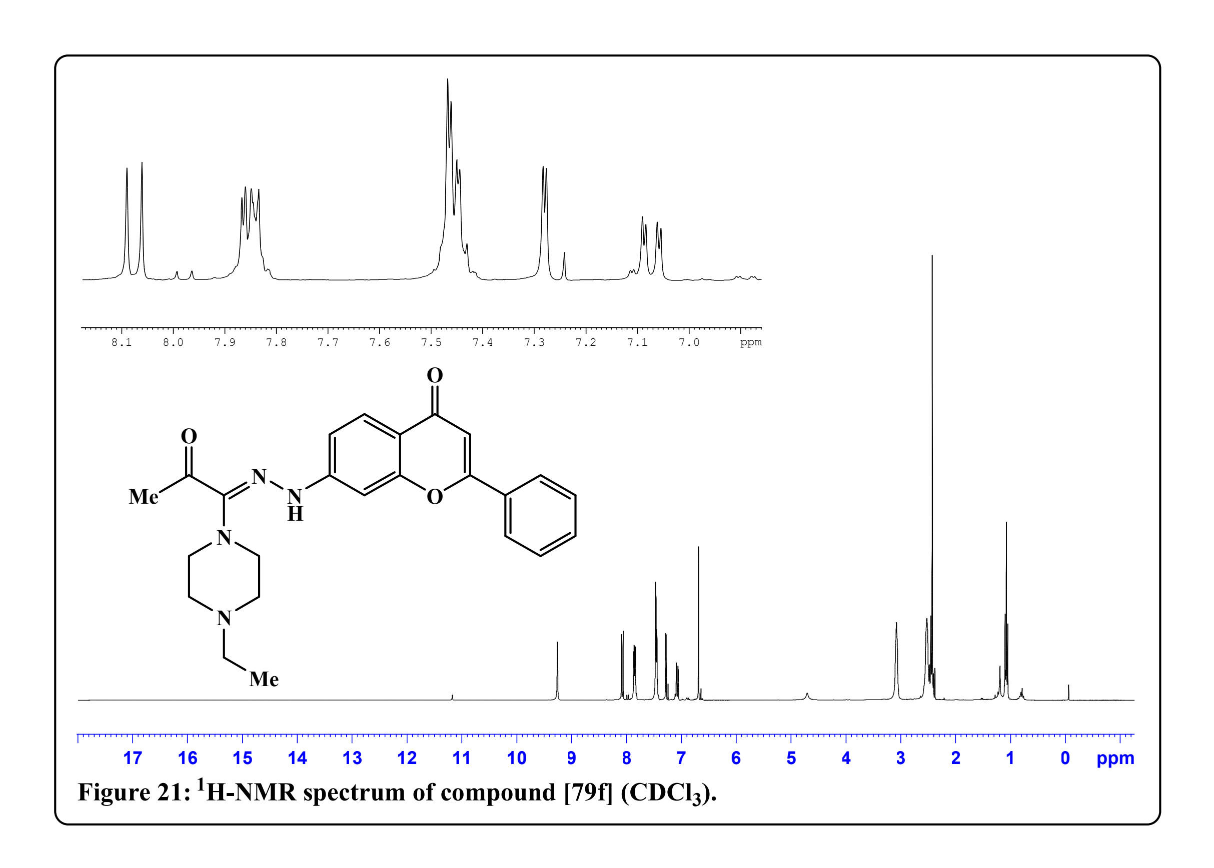


Figure 21: [1]H-NMR spectrum of compound [79f] ($CDCl_3$).

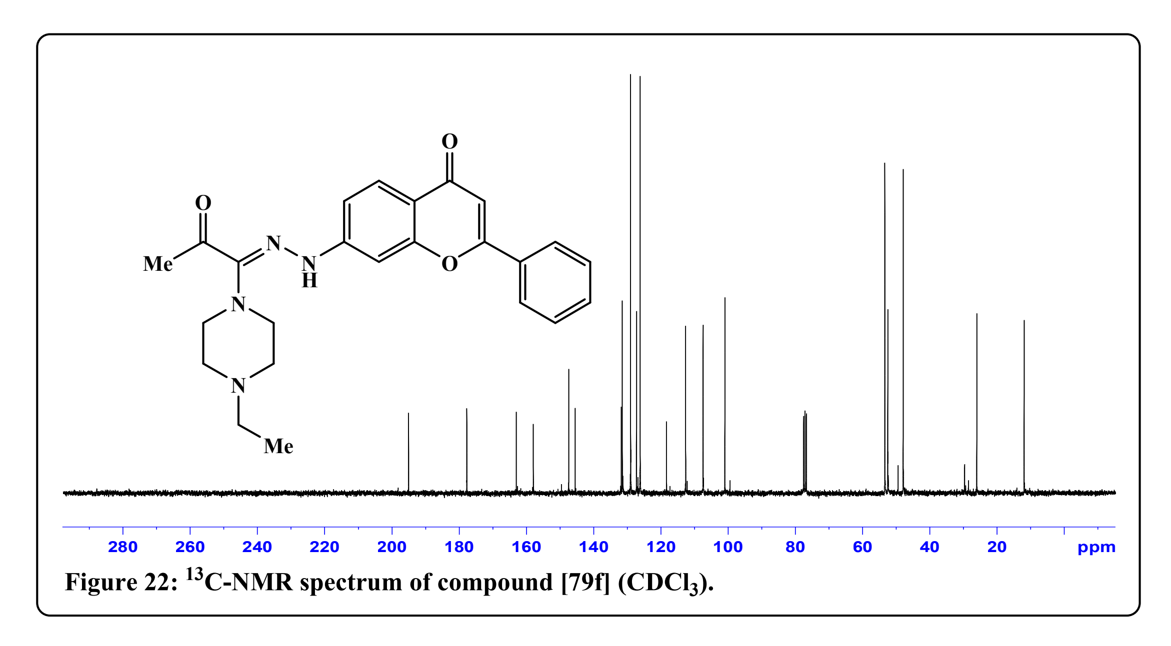


Figure 22: [13]C-NMR spectrum of compound [79f] ($CDCl_3$).

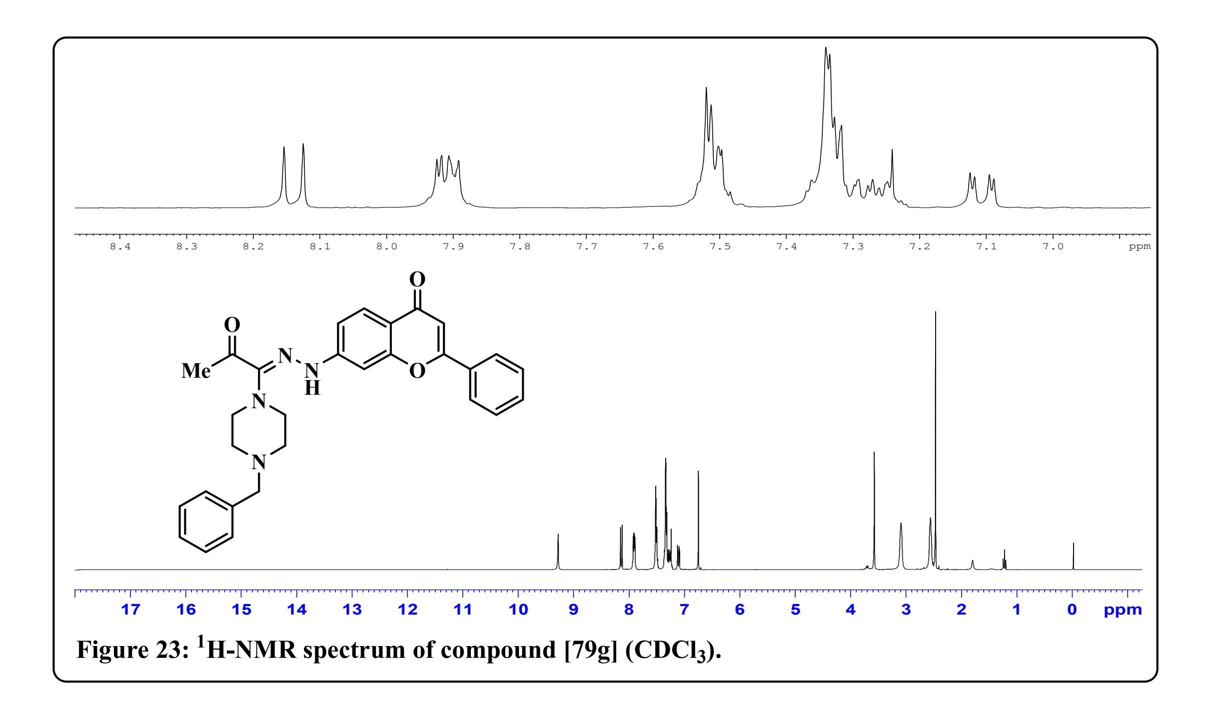


Figure 23: ^{1}H-NMR spectrum of compound [79g] ($CDCl_3$).

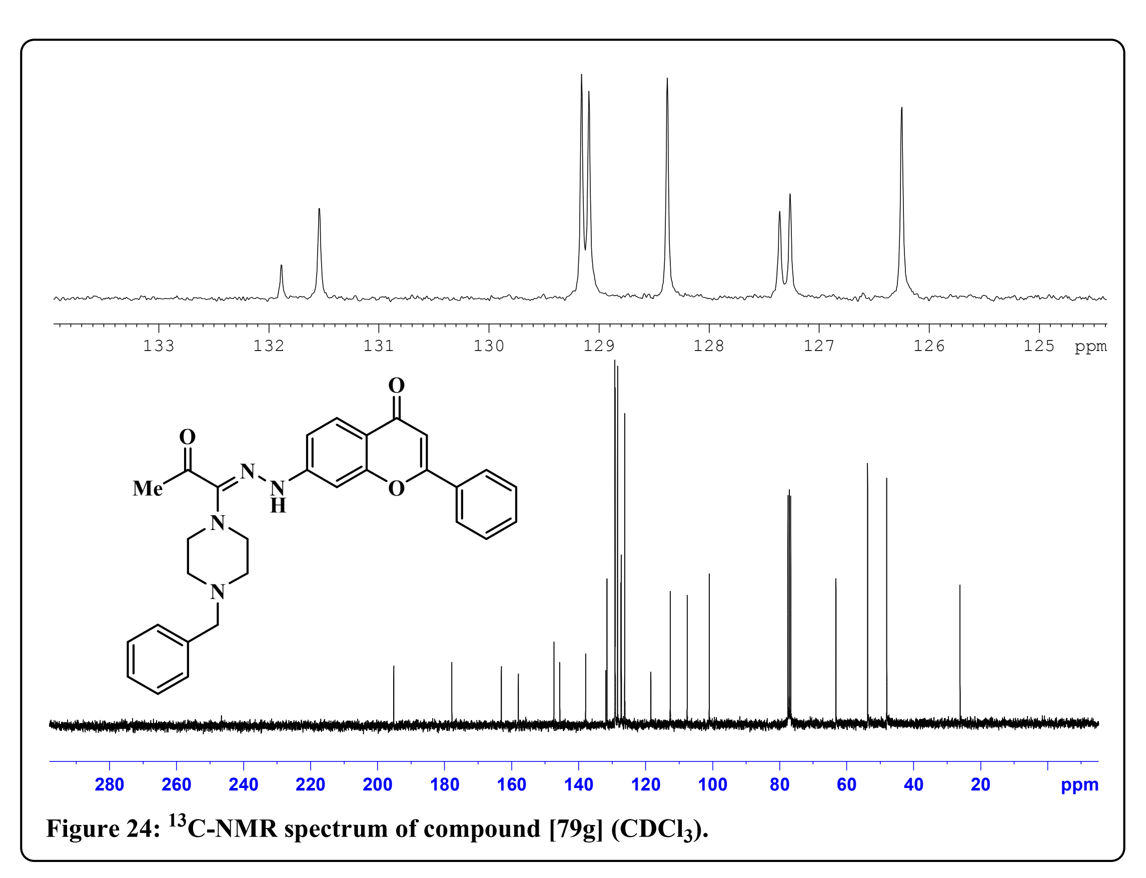


Figure 24: ^{13}C-NMR spectrum of compound [79g] ($CDCl_3$).

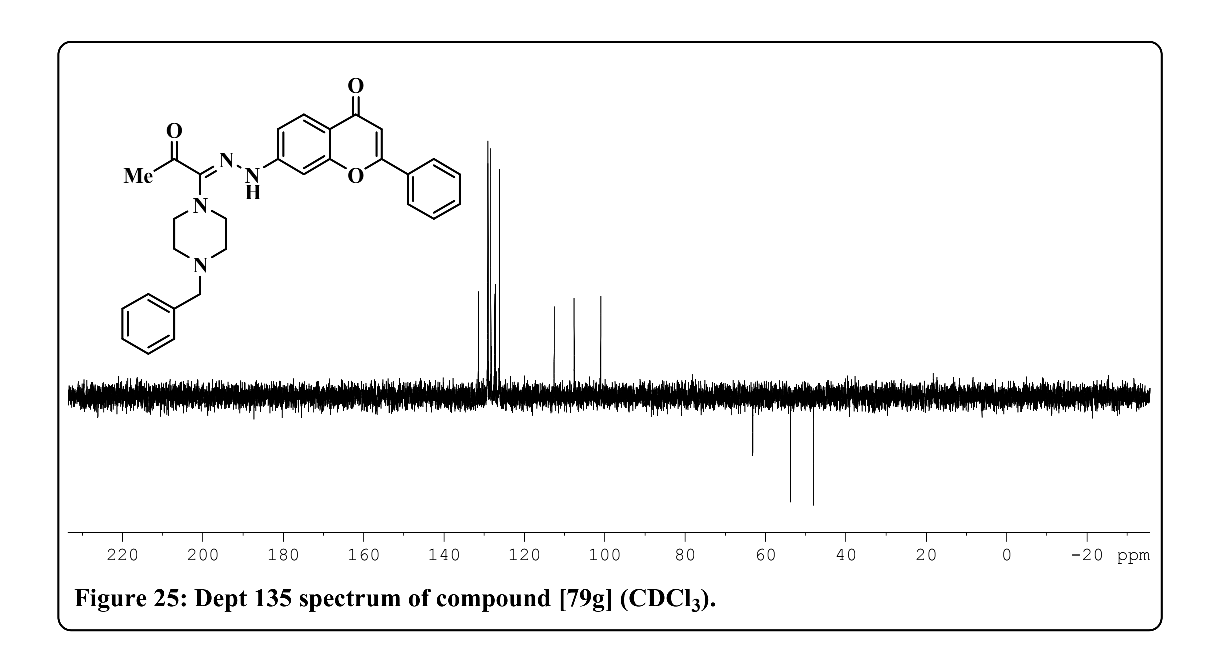


Figure 25: Dept 135 spectrum of compound [79g] ($CDCl_3$).

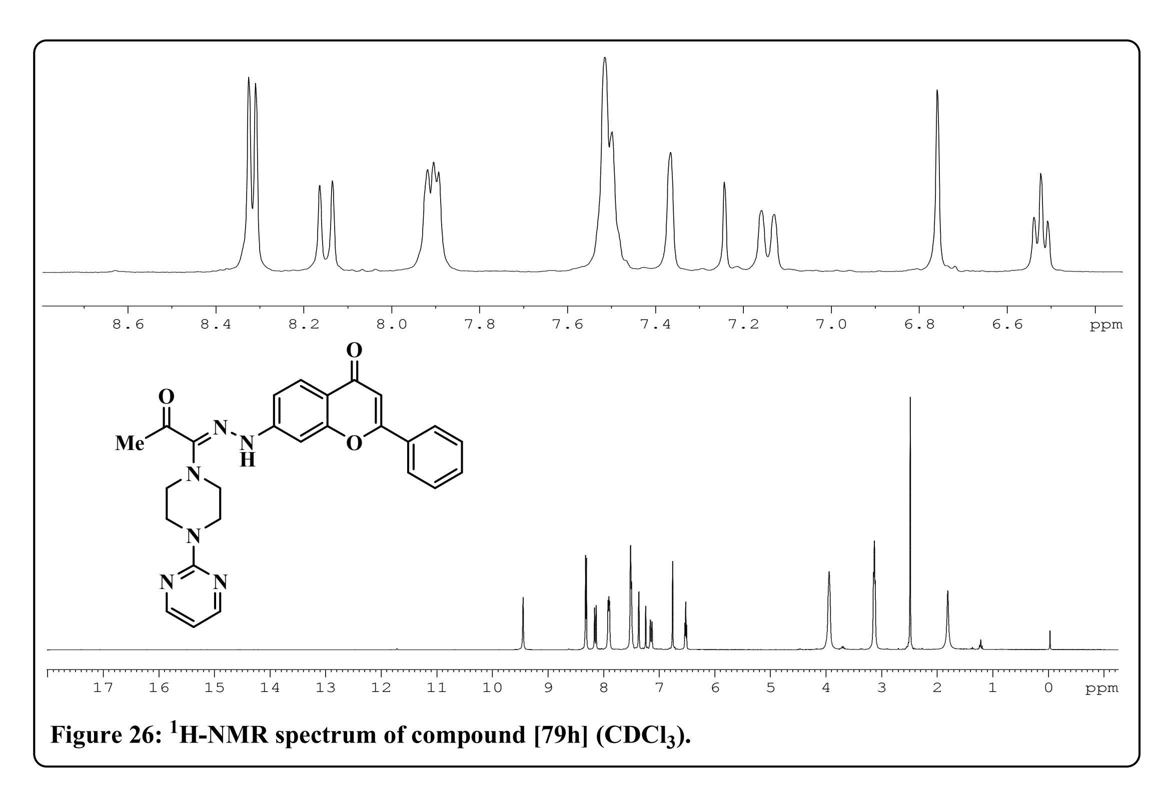


Figure 26: [1]H-NMR spectrum of compound [79h] ($CDCl_3$).

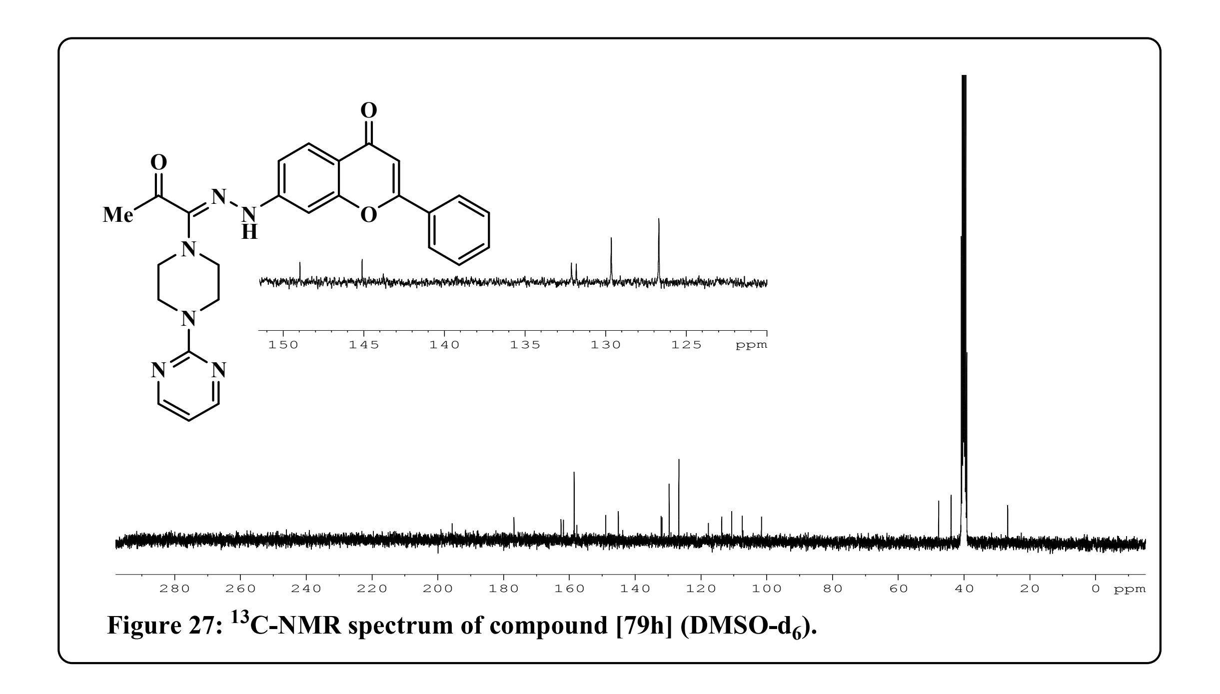


Figure 27: ^{13}C-NMR spectrum of compound [79h] (DMSO-d_6).

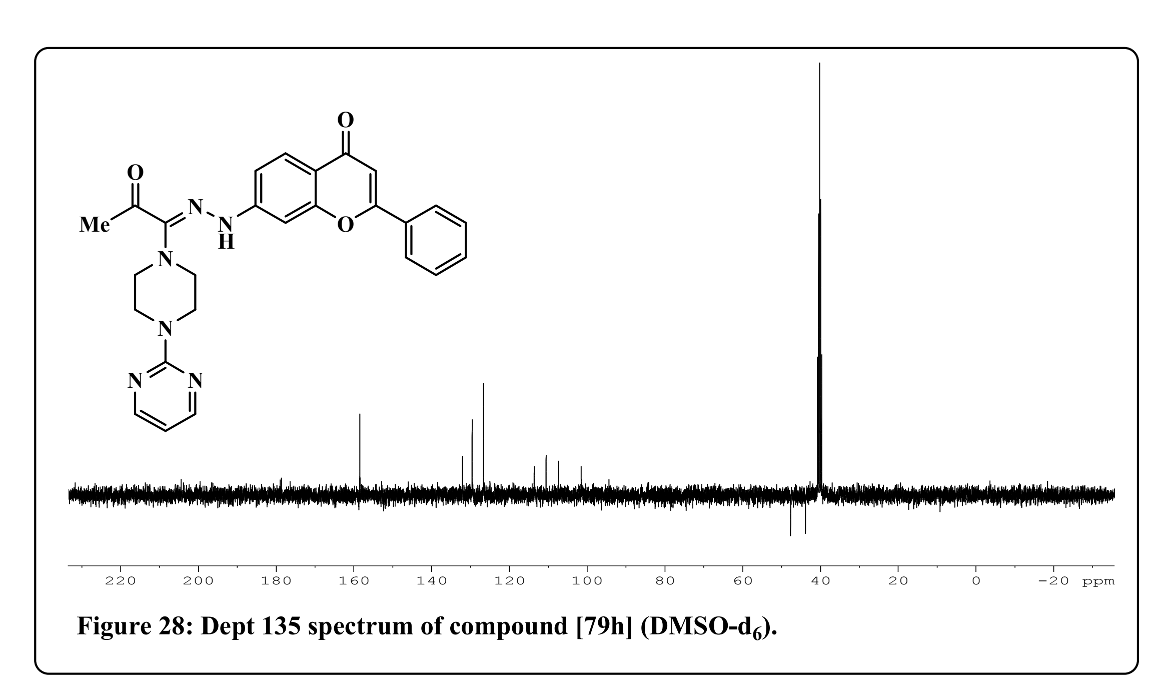


Figure 28: Dept 135 spectrum of compound [79h] (DMSO-d_6).

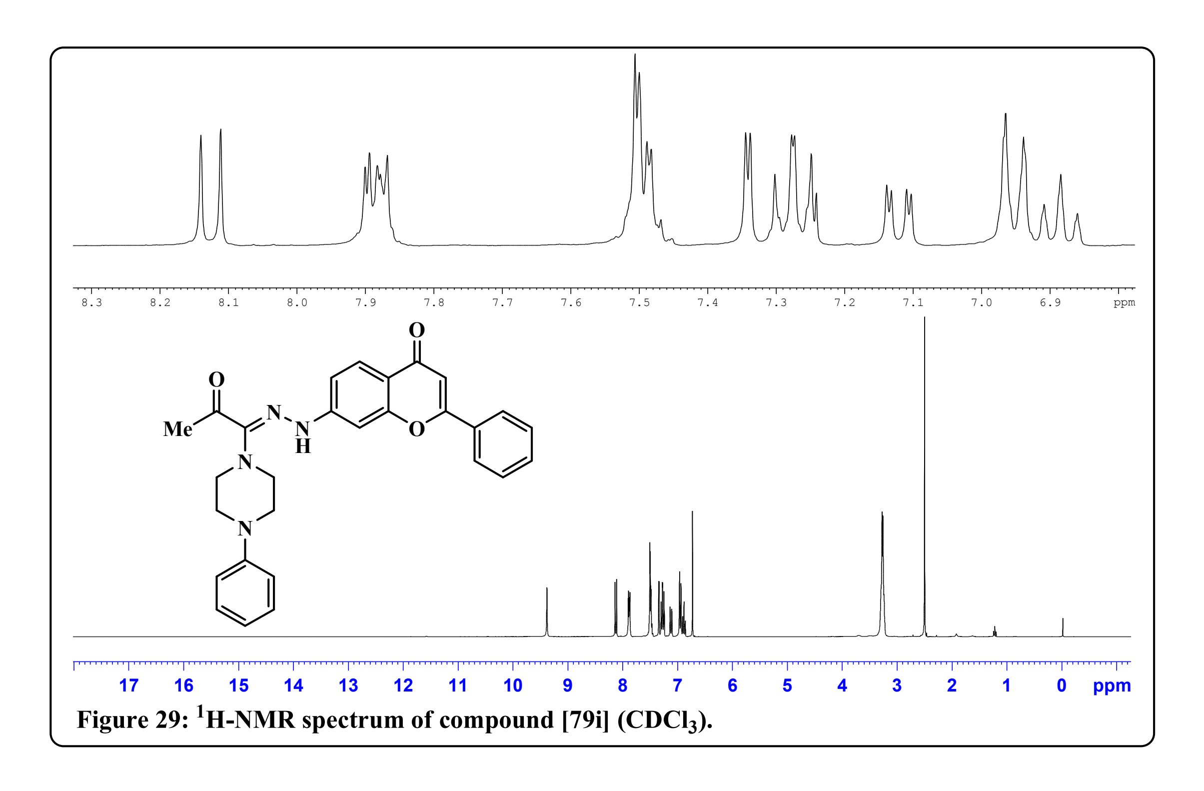


Figure 29: ^{1}H-NMR spectrum of compound [79i] ($CDCl_3$).

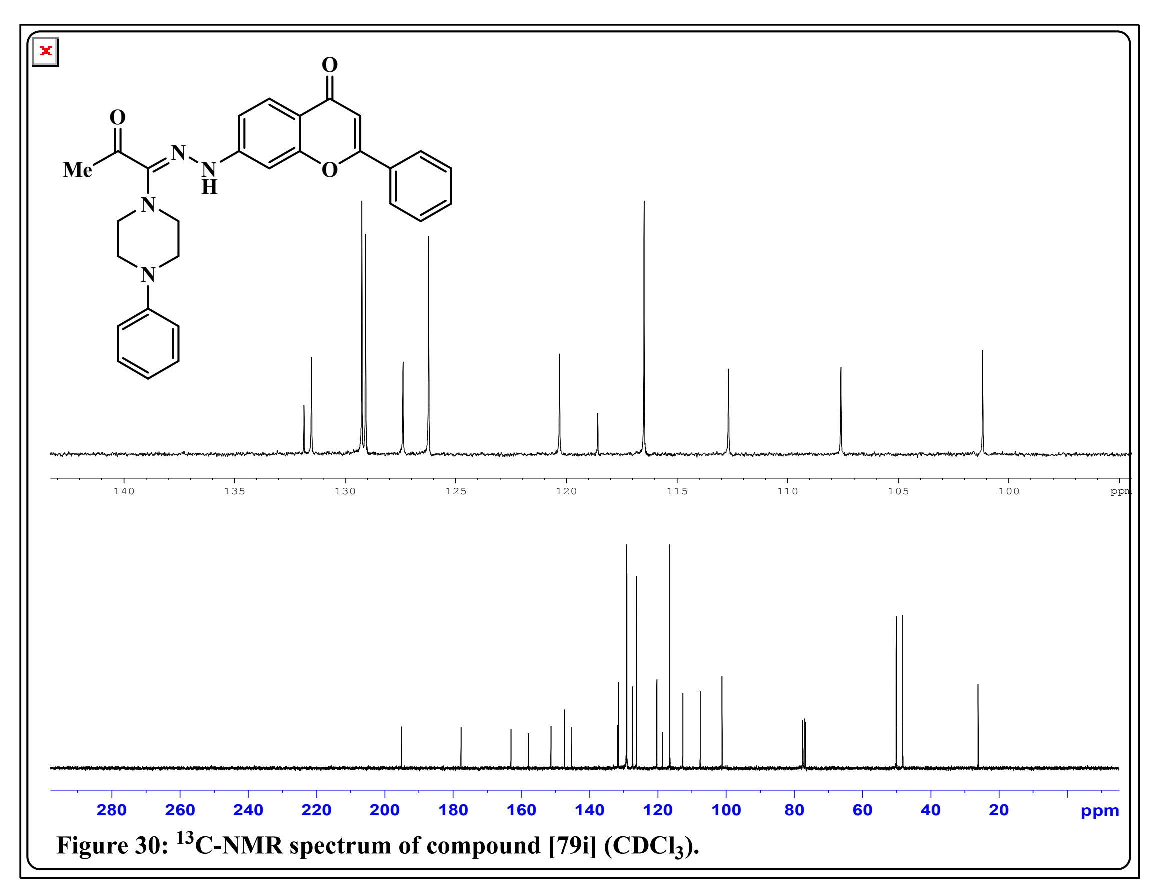


Figure 30: ^{13}C-NMR spectrum of compound [79i] ($CDCl_3$).

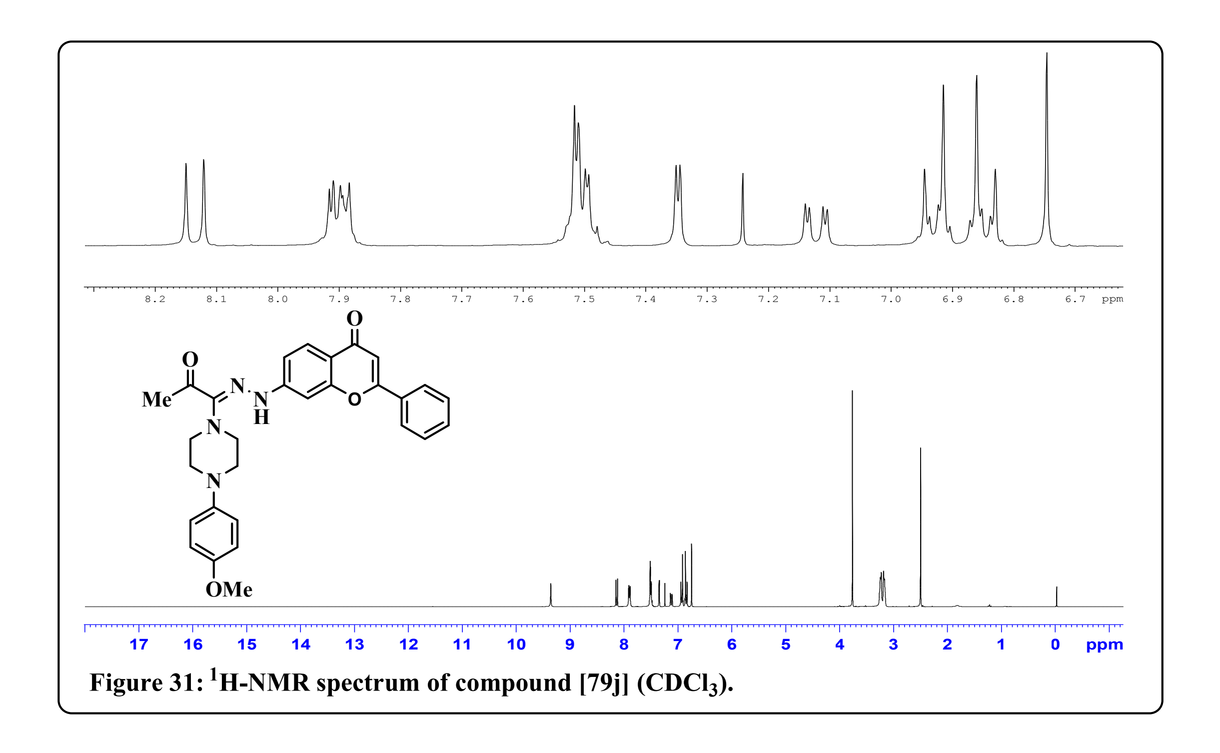


Figure 31: ^{1}H-NMR spectrum of compound [79j] ($CDCl_3$).

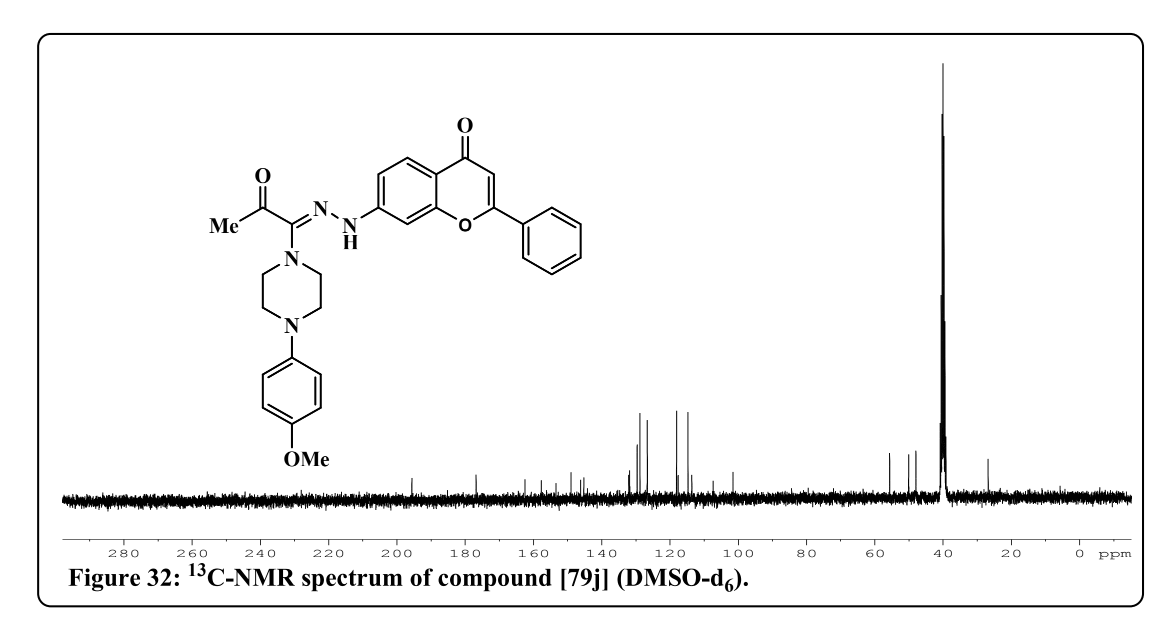


Figure 32: ^{13}C-NMR spectrum of compound [79j] (DMSO-d_6).

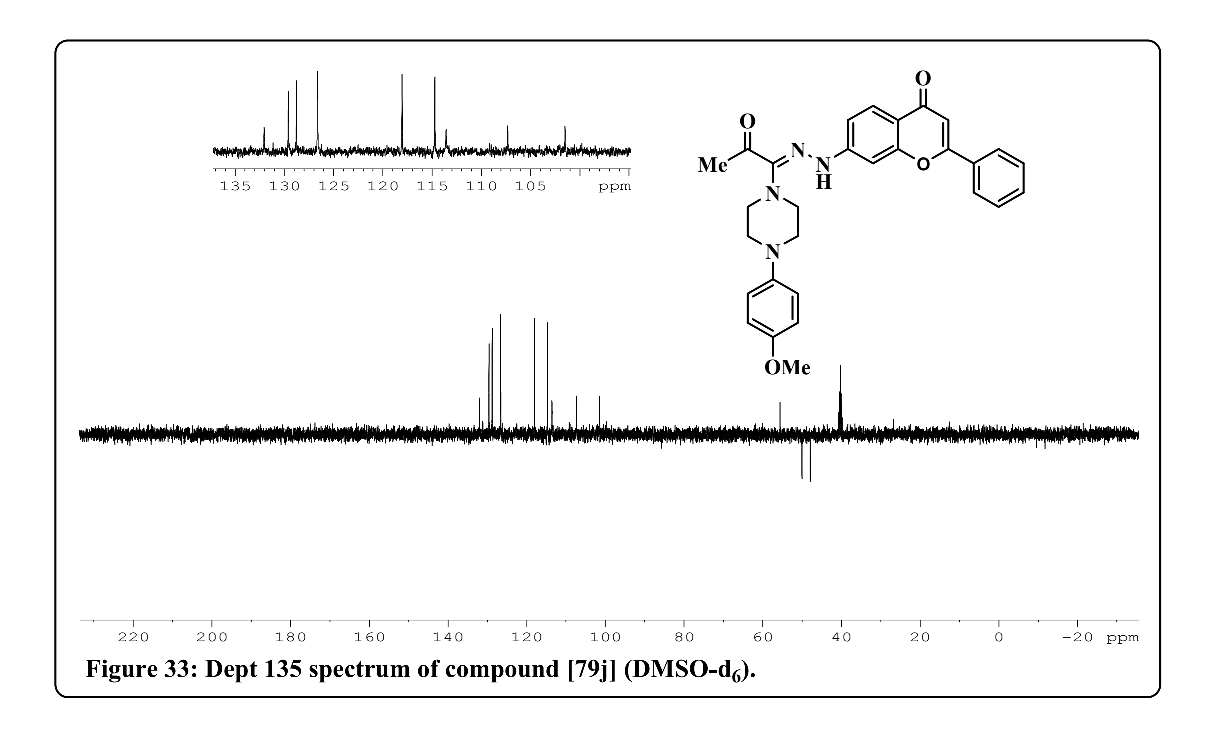


Figure 33: Dept 135 spectrum of compound [79j] (DMSO-d_6).

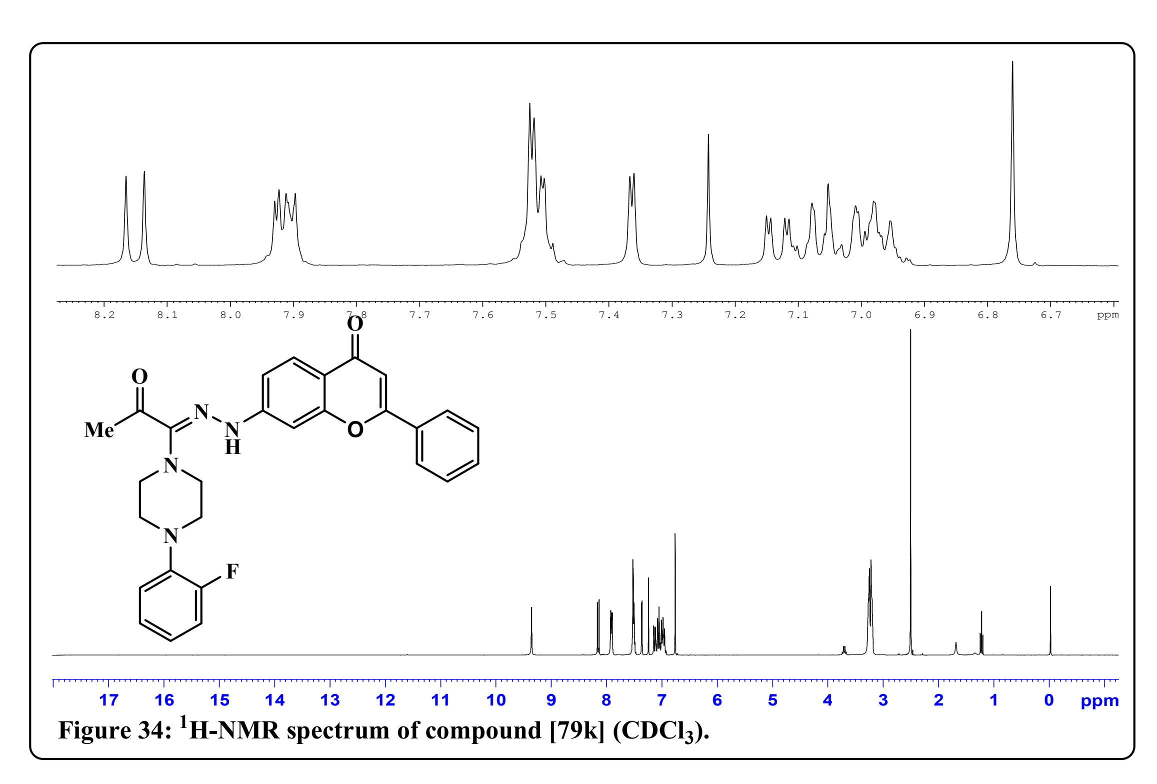


Figure 34: ^{1}H-NMR spectrum of compound [79k] ($CDCl_3$).

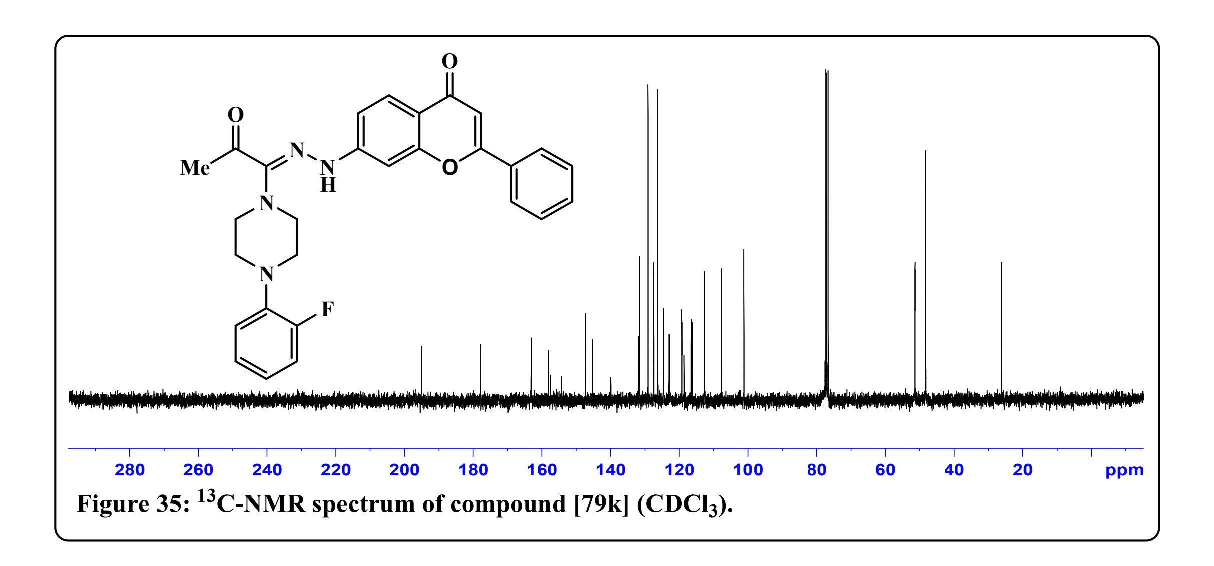


Figure 35: ^{13}C-NMR spectrum of compound [79k] ($CDCl_3$).

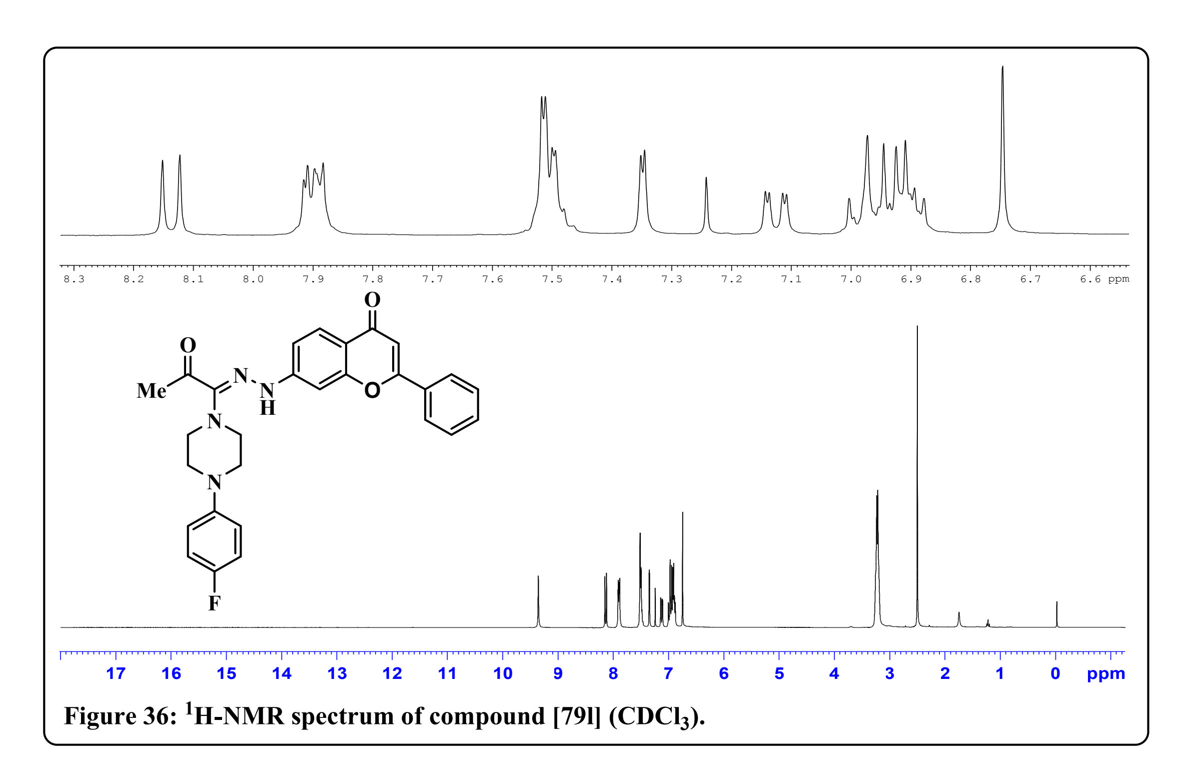


Figure 36: ^{1}H-NMR spectrum of compound [79l] ($CDCl_3$).

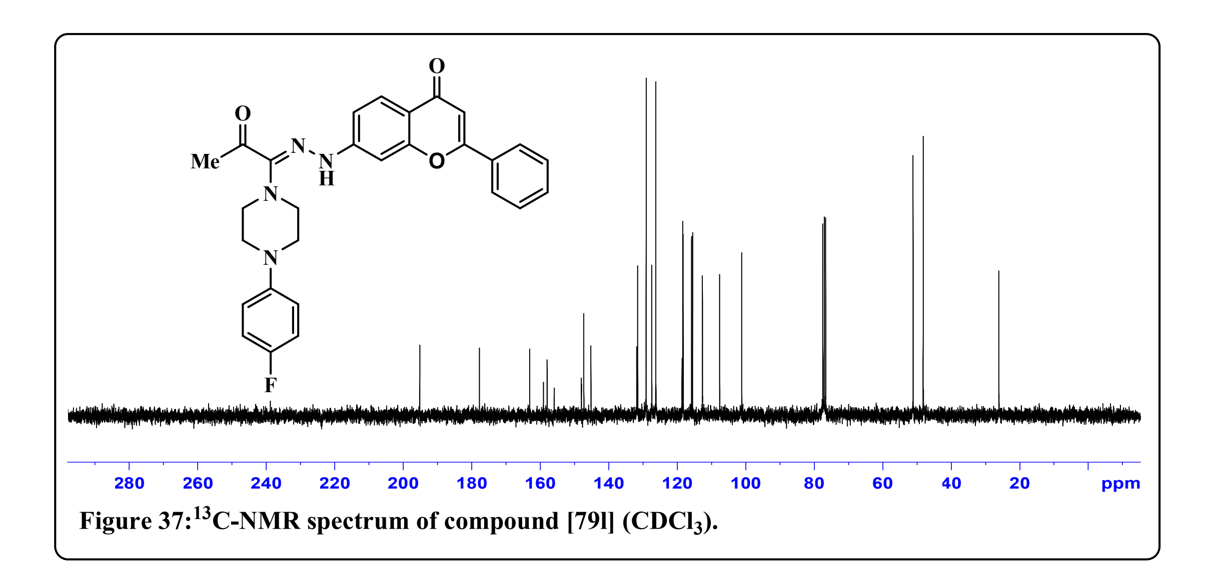


Figure 37: ^{13}C-NMR spectrum of compound [79l] ($CDCl_3$).

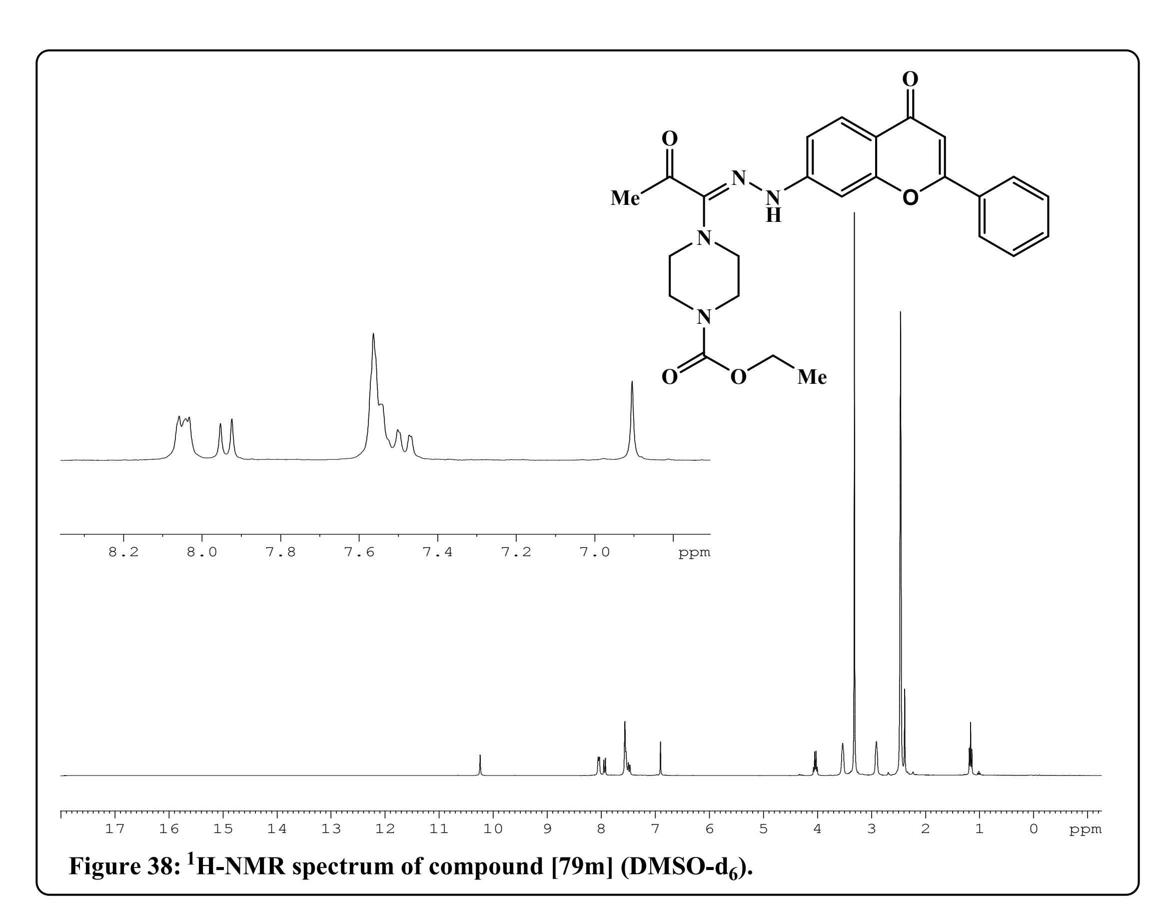


Figure 38: ^{1}H-NMR spectrum of compound [79m] (DMSO-d_6).

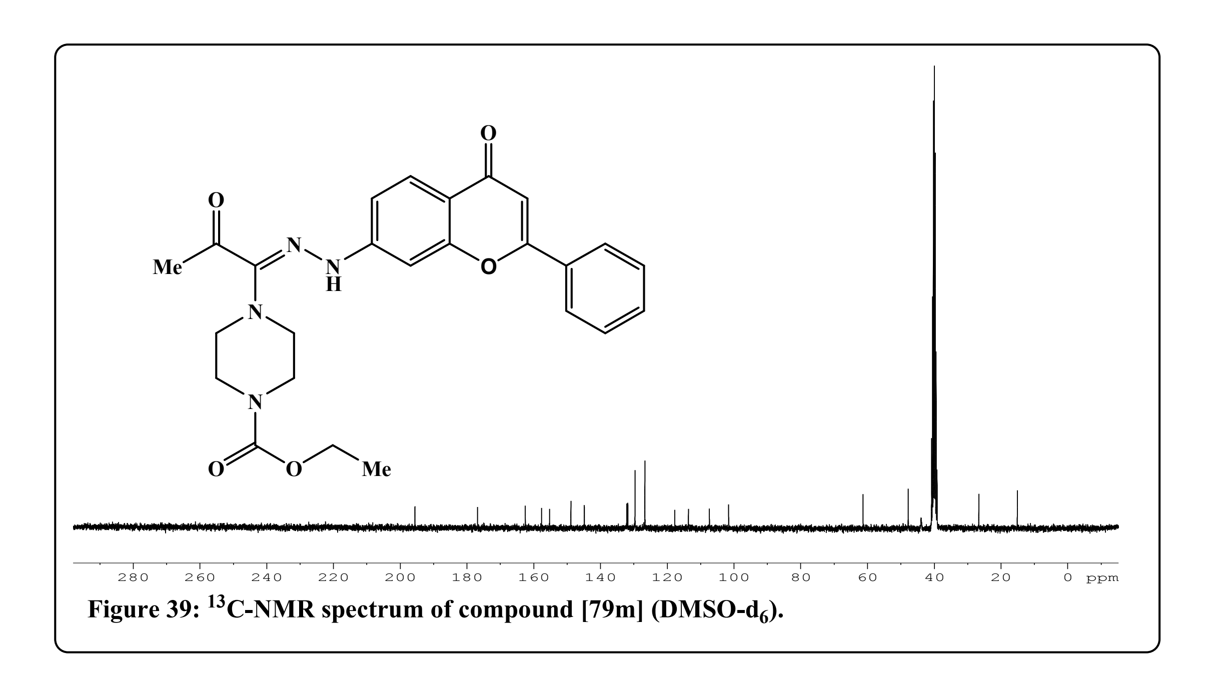


Figure 39: ^{13}C-NMR spectrum of compound [79m] (DMSO-d_6).

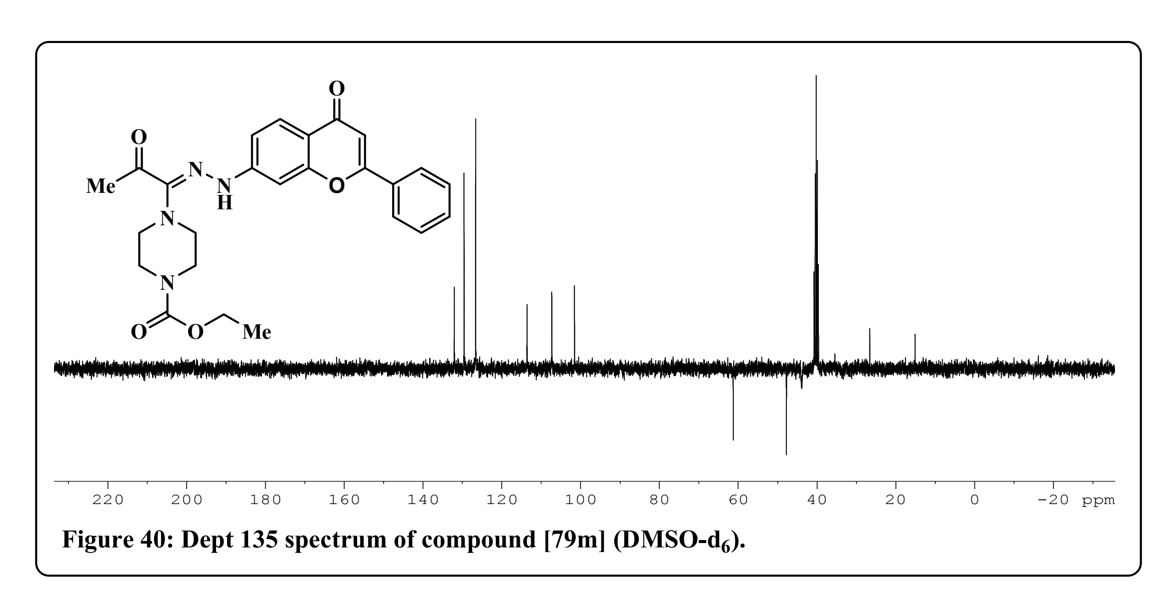


Figure 40: Dept 135 spectrum of compound [79m] (DMSO-d_6).

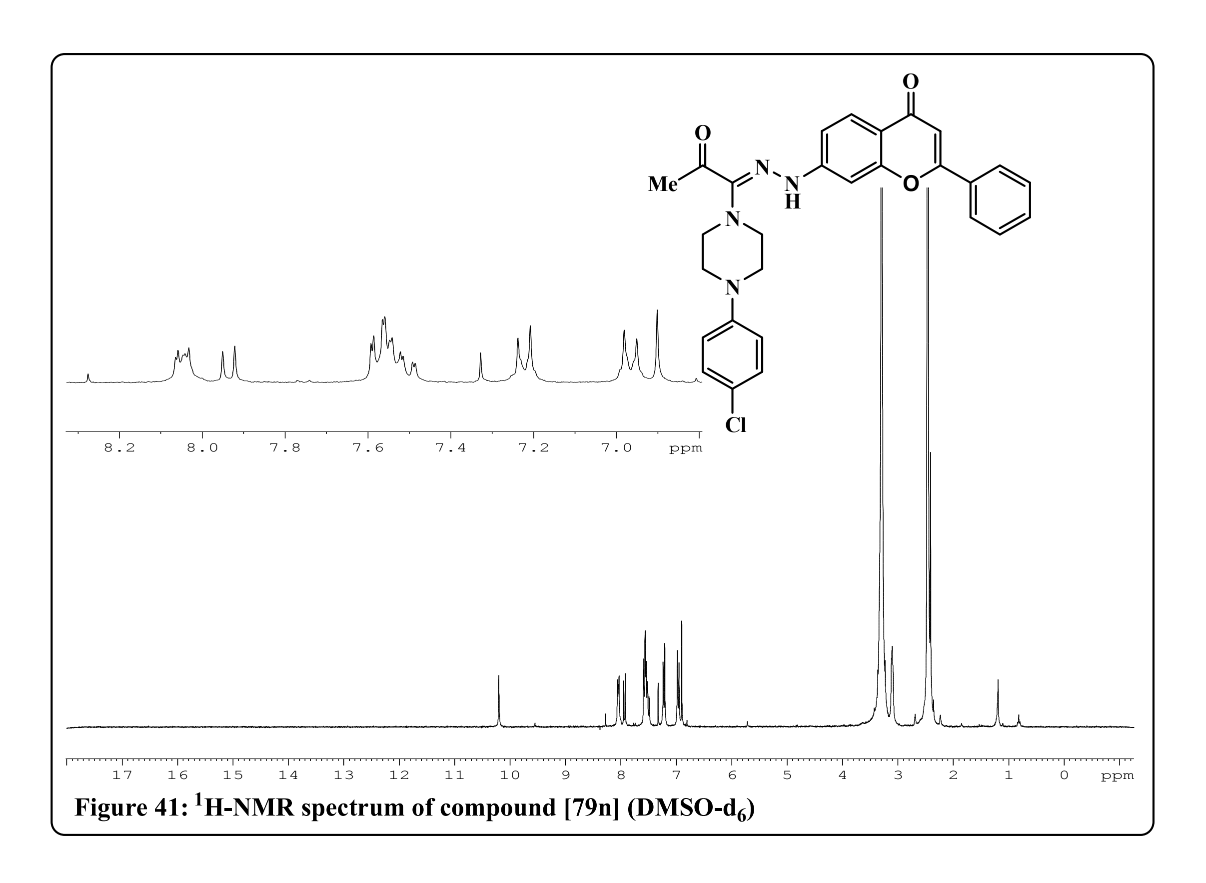


Figure 41: ^{1}H-NMR spectrum of compound [79n] (DMSO-d_6)

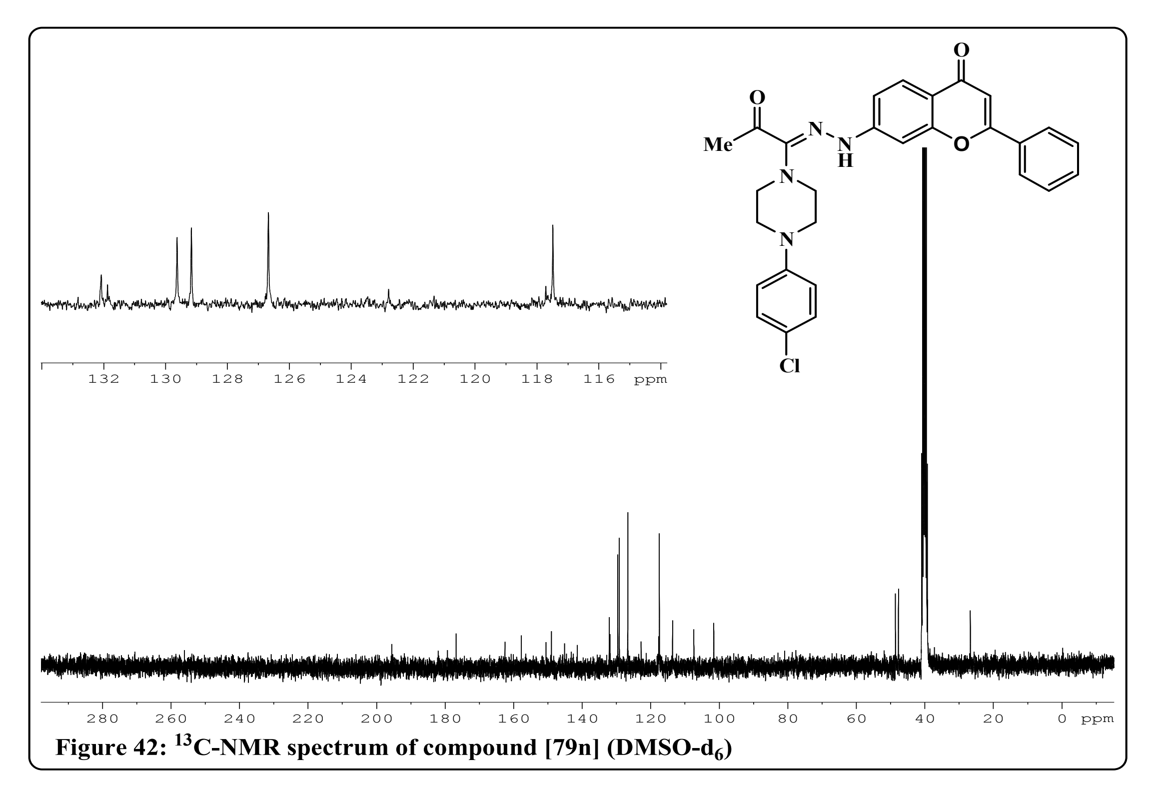


Figure 42: ^{13}C-NMR spectrum of compound [79n] (DMSO-d_6)

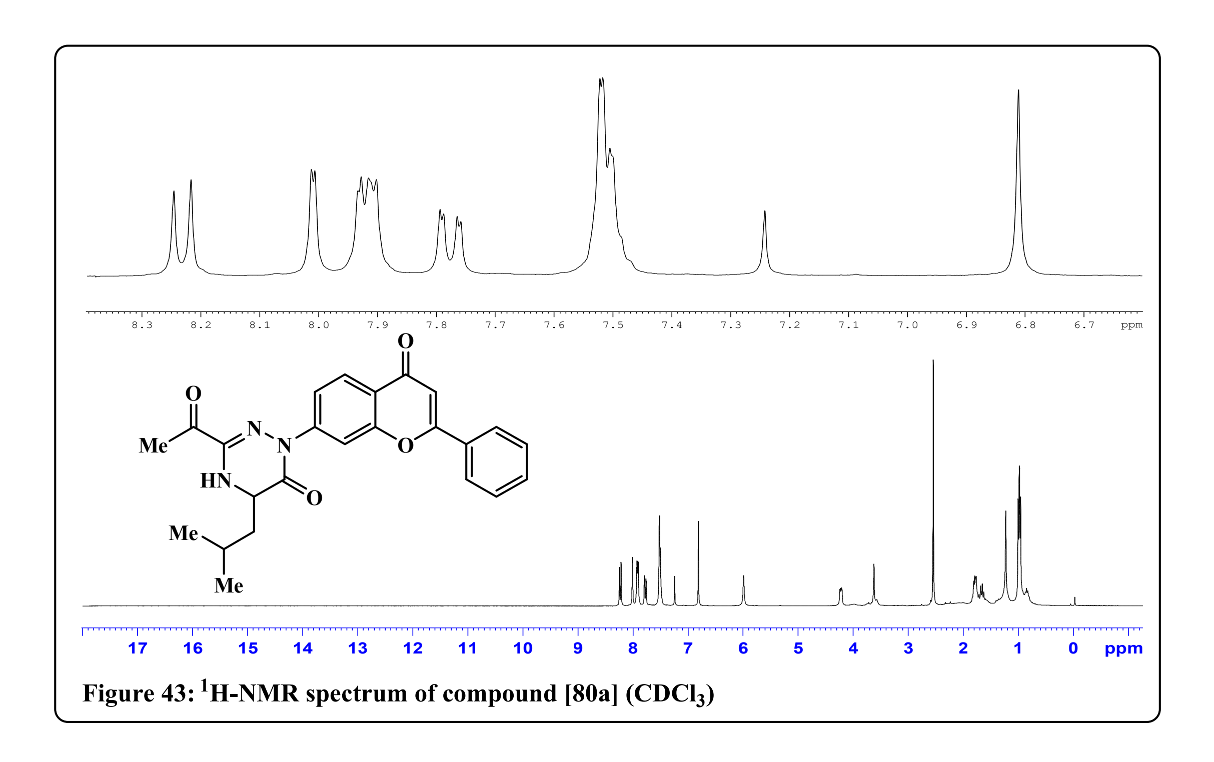


Figure 43: ^{1}H-NMR spectrum of compound [80a] ($CDCl_3$)

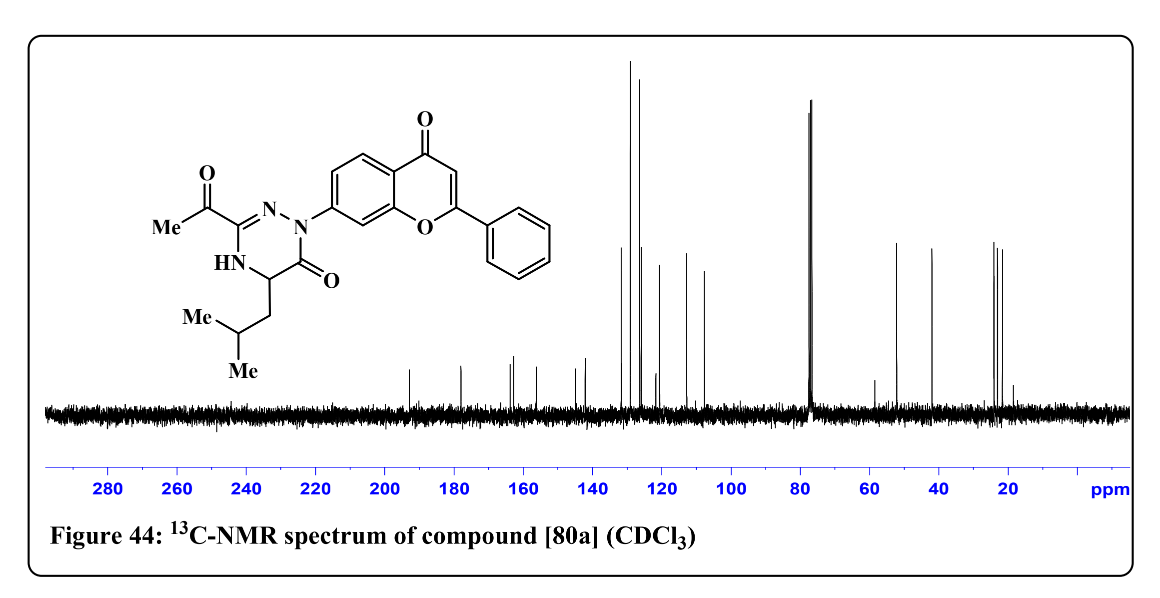


Figure 44: ^{13}C-NMR spectrum of compound [80a] ($CDCl_3$)

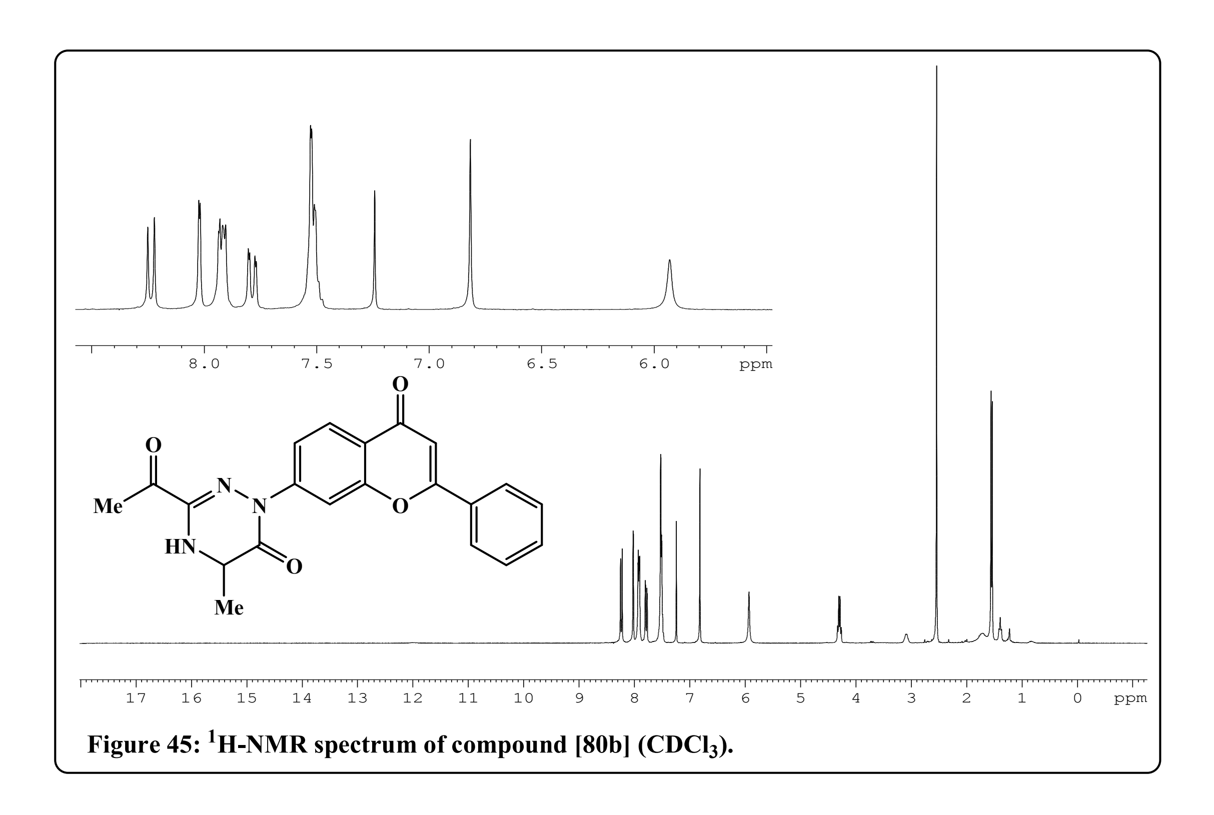


Figure 45: ^{1}H-NMR spectrum of compound [80b] ($CDCl_3$).

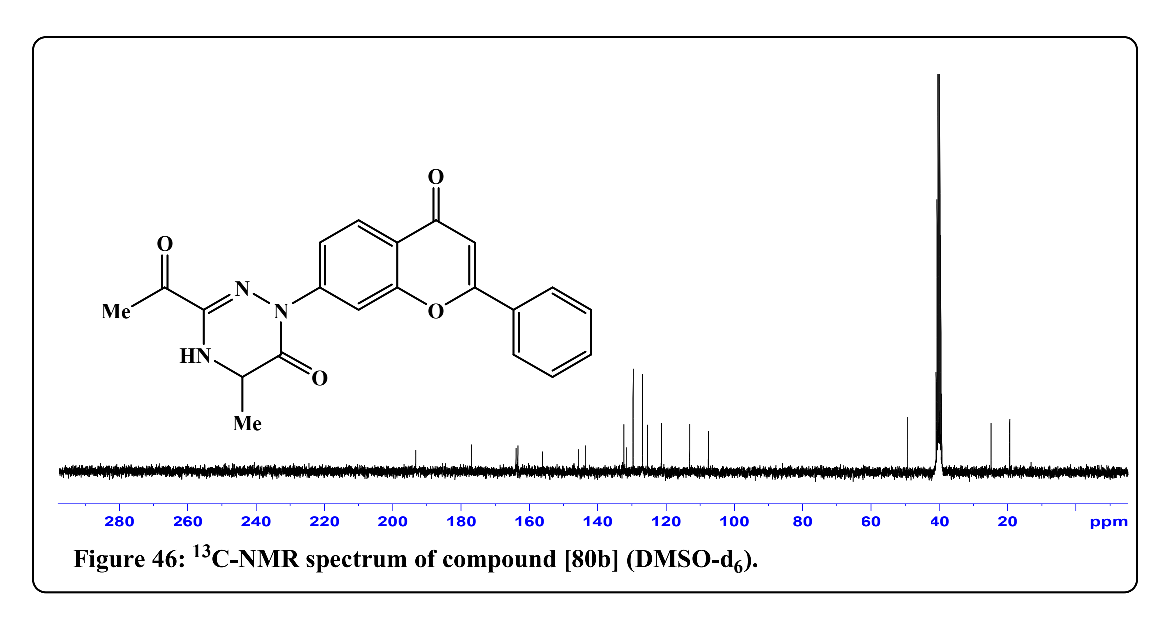


Figure 46: ^{13}C-NMR spectrum of compound [80b] (DMSO-d_6).

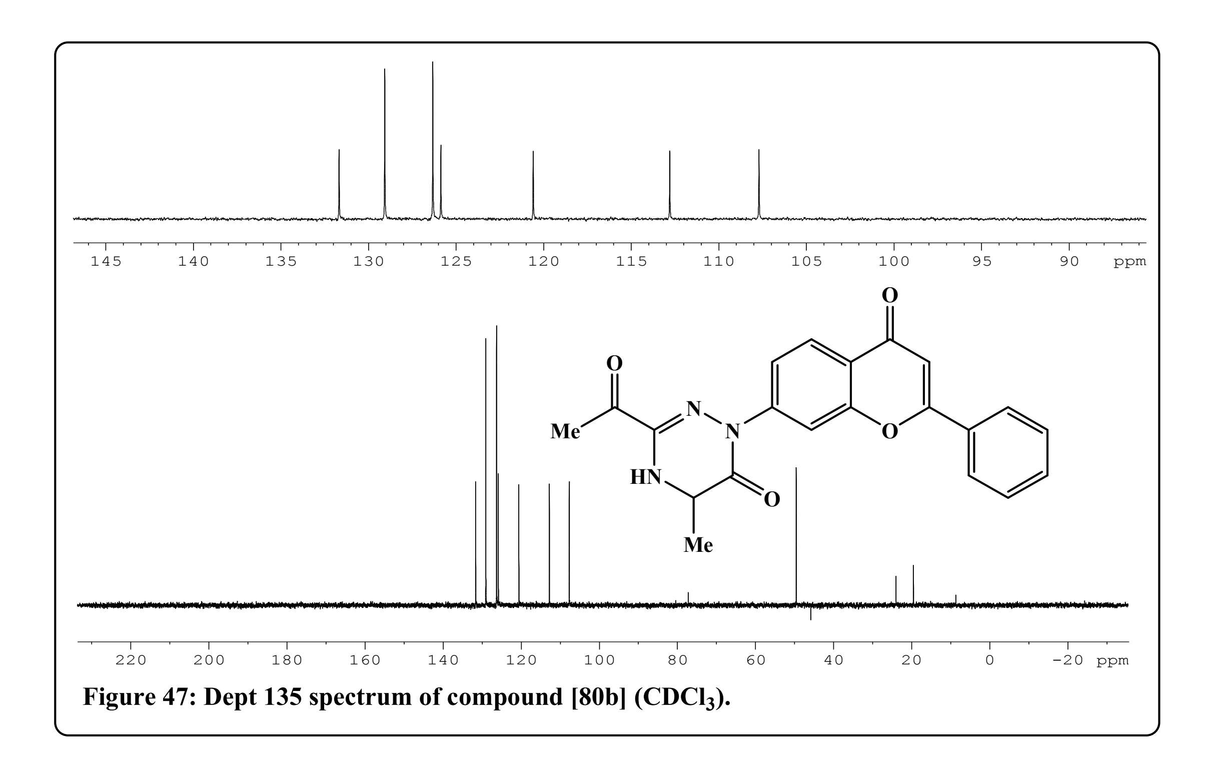


Figure 47: Dept 135 spectrum of compound [80b] ($CDCl_3$).

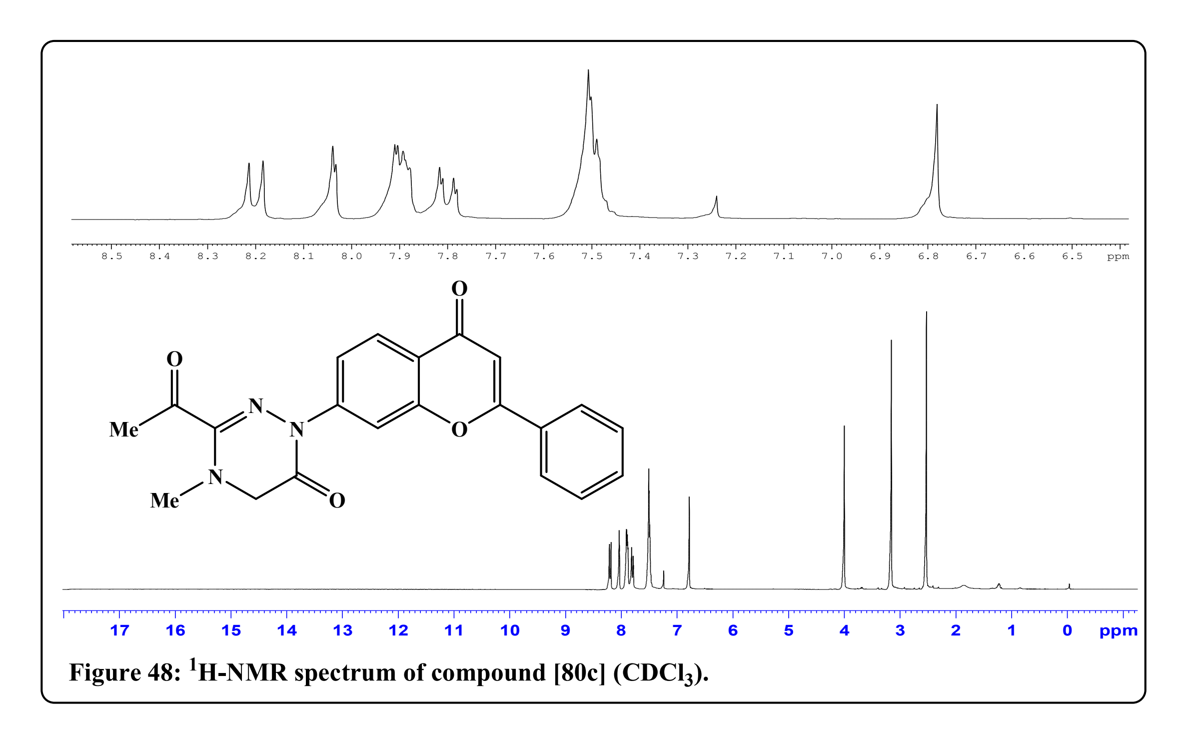


Figure 48: ^{1}H-NMR spectrum of compound [80c] ($CDCl_3$).

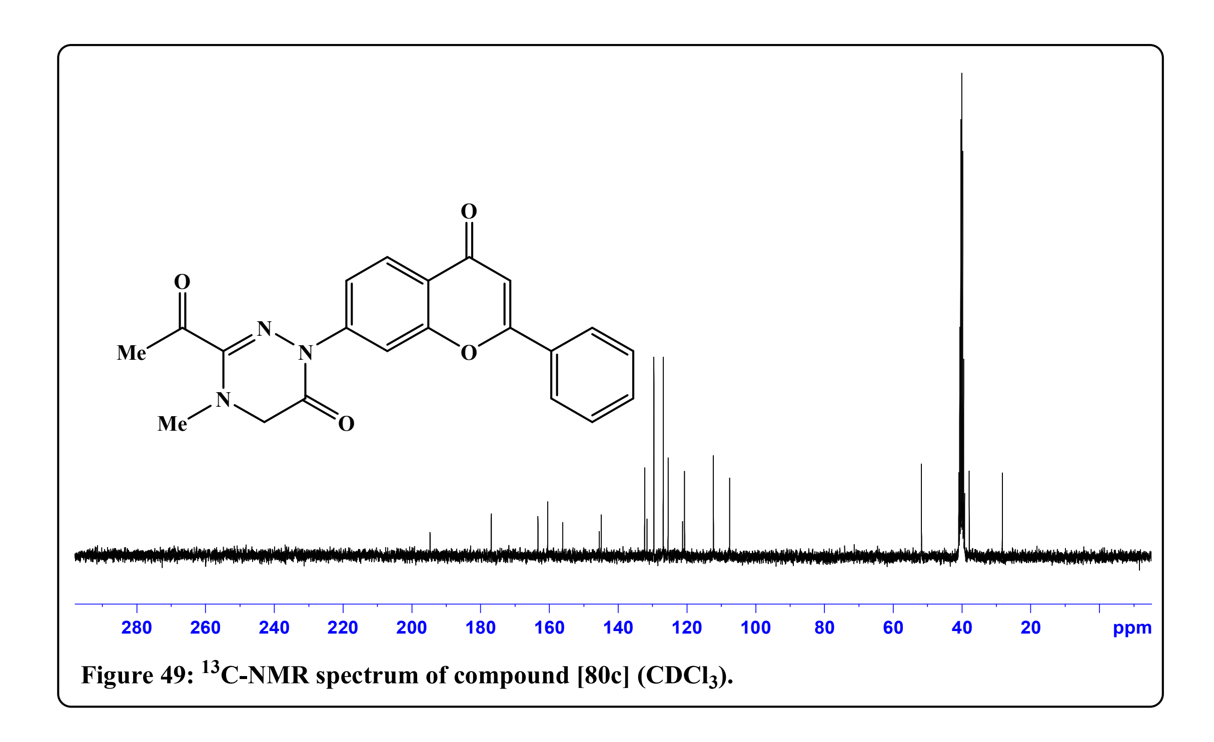


Figure 49: ^{13}C-NMR spectrum of compound [80c] ($CDCl_3$).

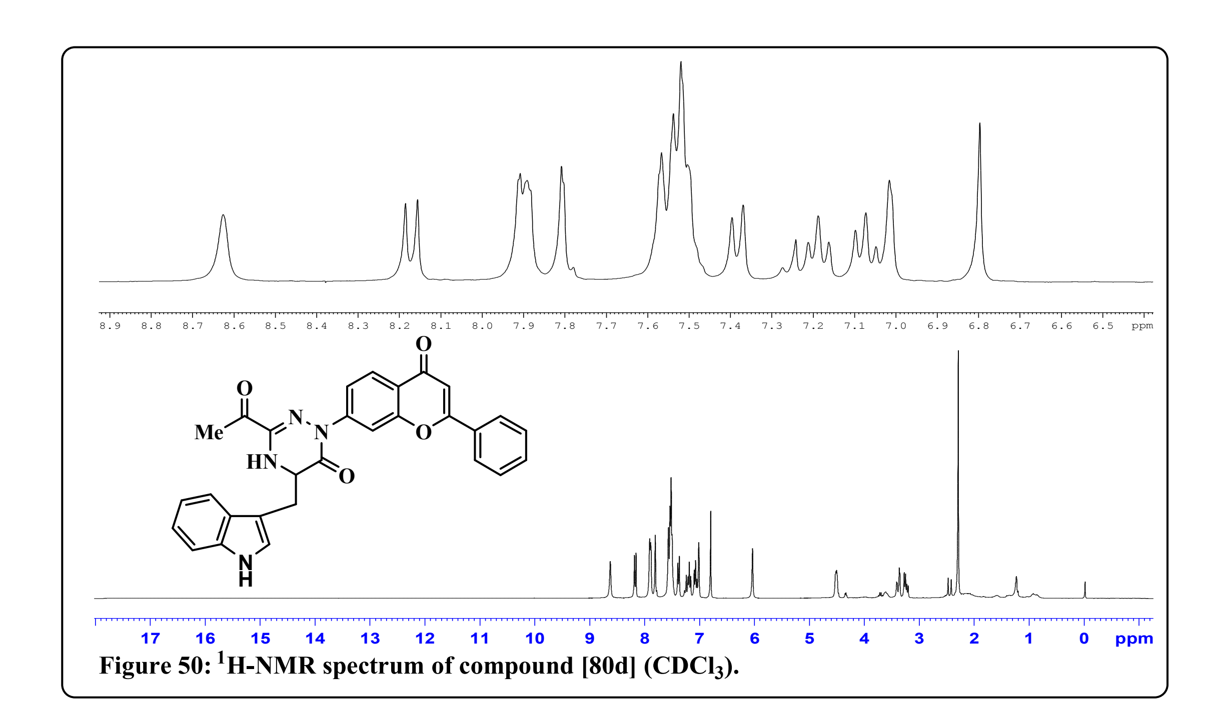


Figure 50: ^{1}H-NMR spectrum of compound [80d] ($CDCl_3$).

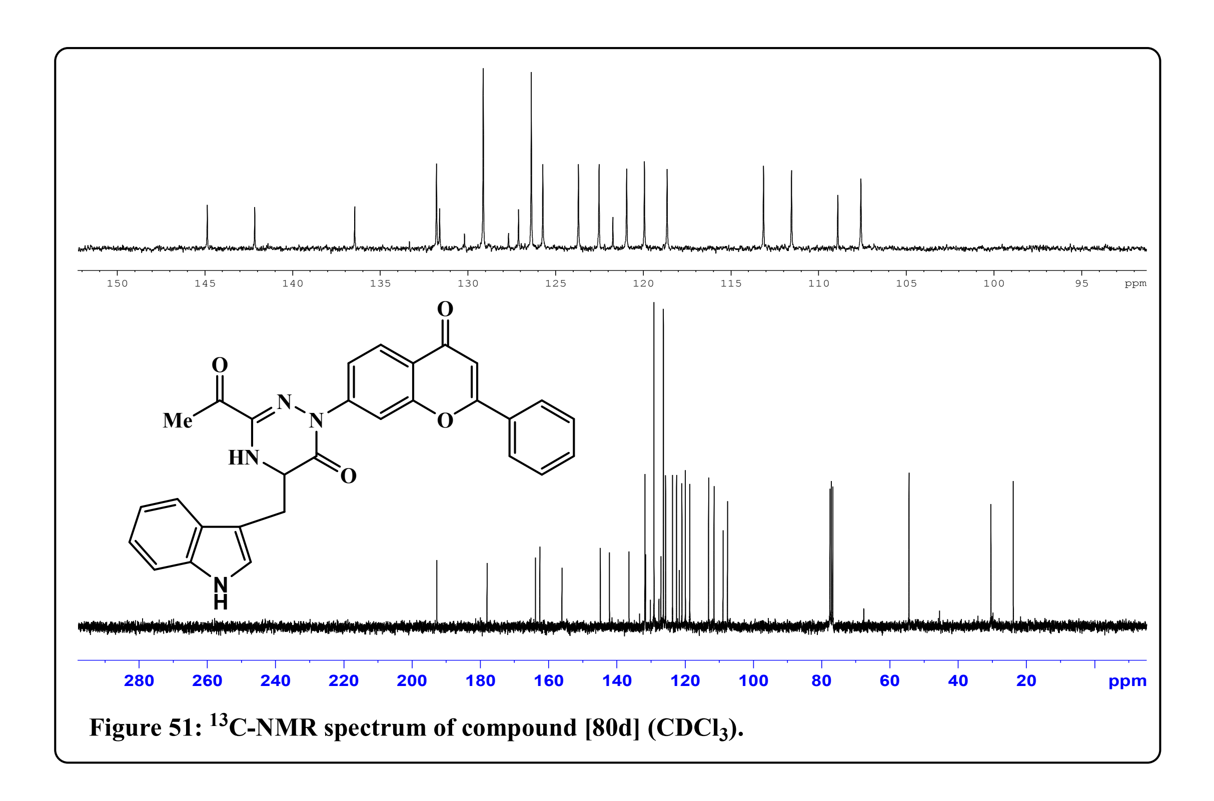


Figure 51: ^{13}C-NMR spectrum of compound [80d] ($CDCl_3$).

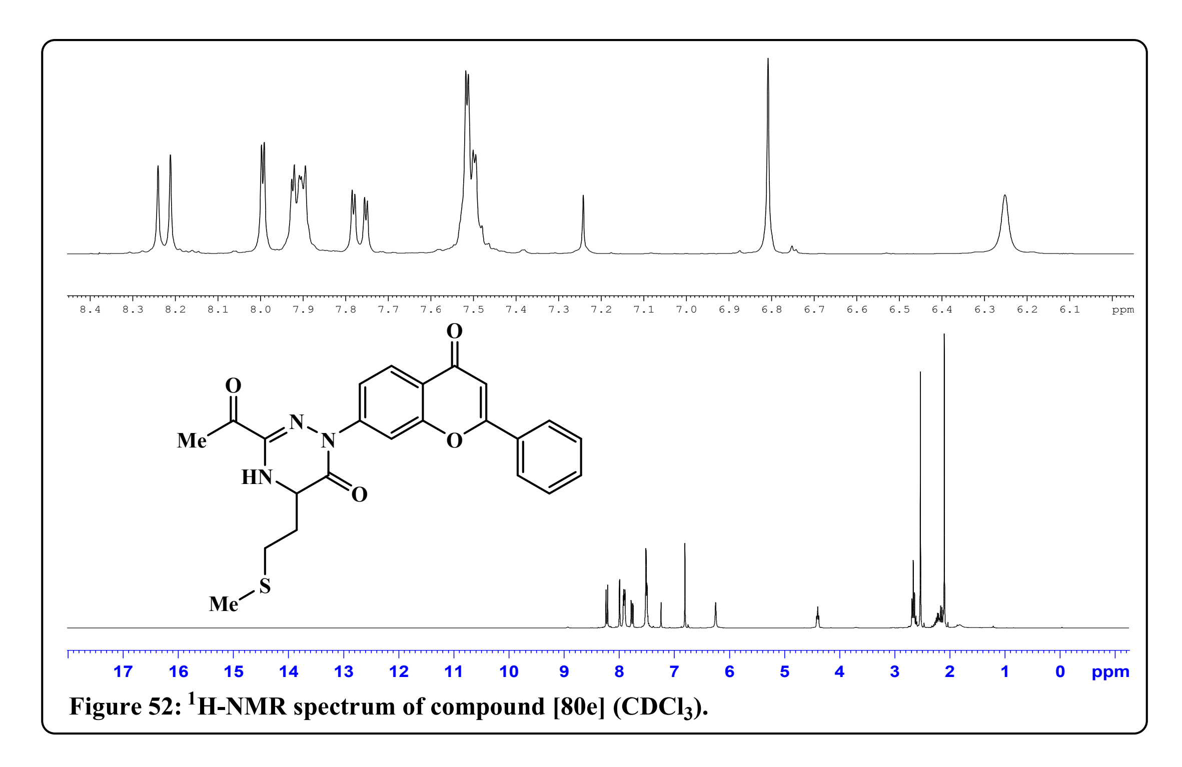


Figure 52: ^{1}H-NMR spectrum of compound [80e] ($CDCl_3$).

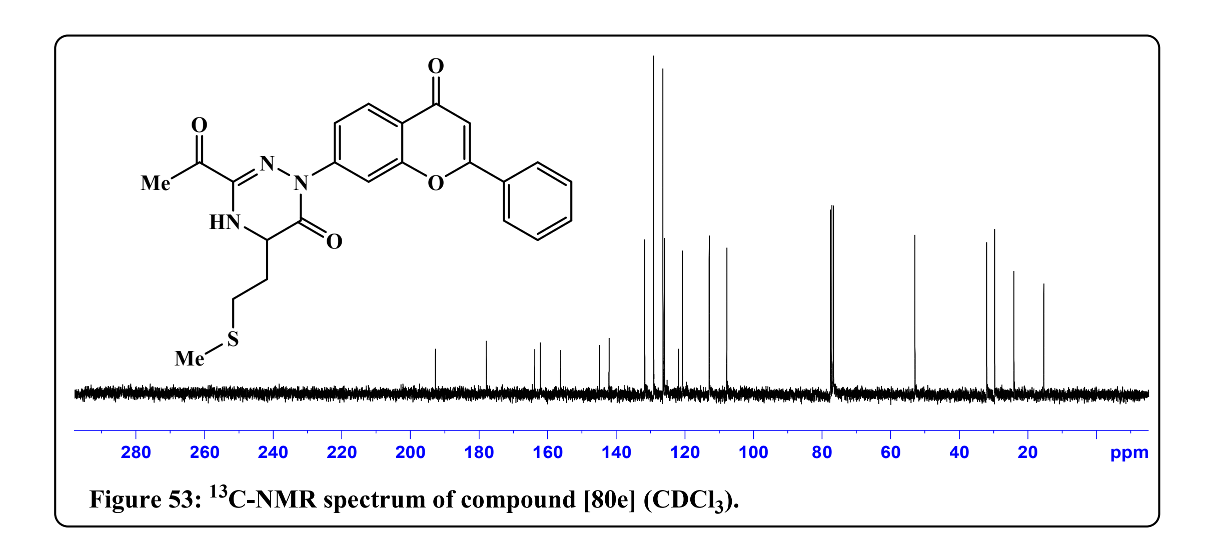


Figure 53: ^{13}C-NMR spectrum of compound [80e] ($CDCl_3$).

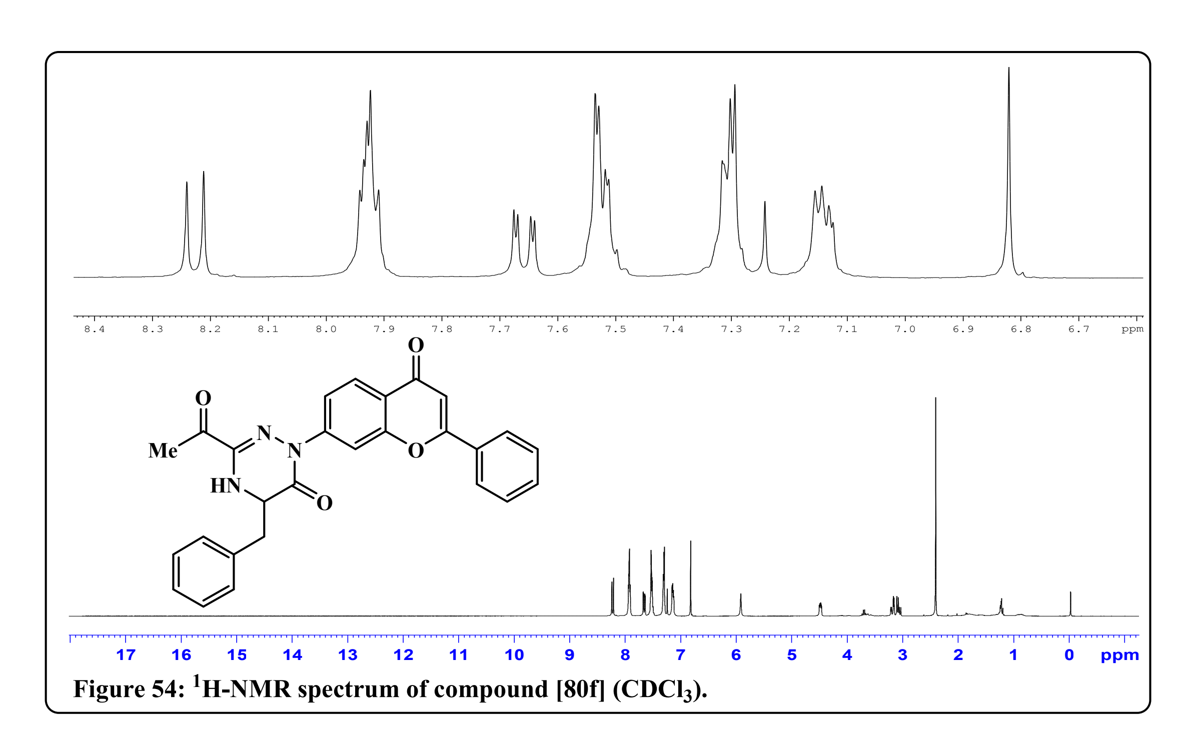


Figure 54: ^{1}H-NMR spectrum of compound [80f] ($CDCl_3$).

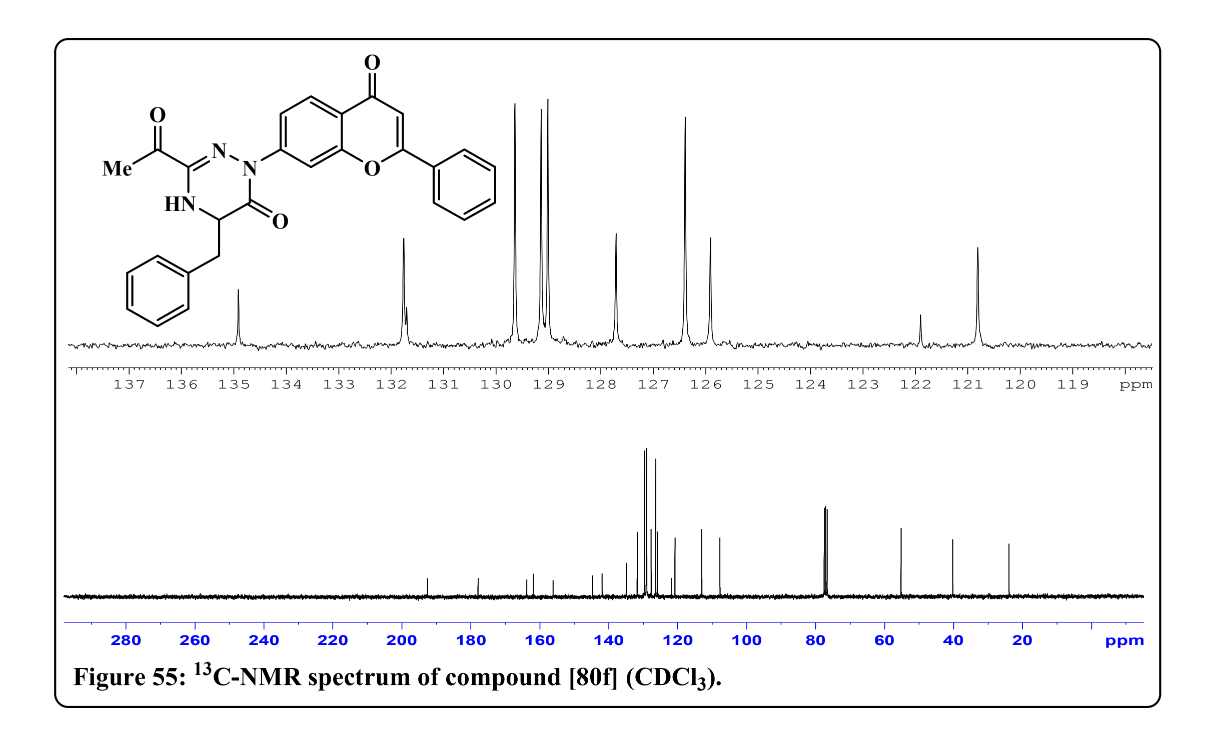


Figure 55: ^{13}C-NMR spectrum of compound [80f] ($CDCl_3$).

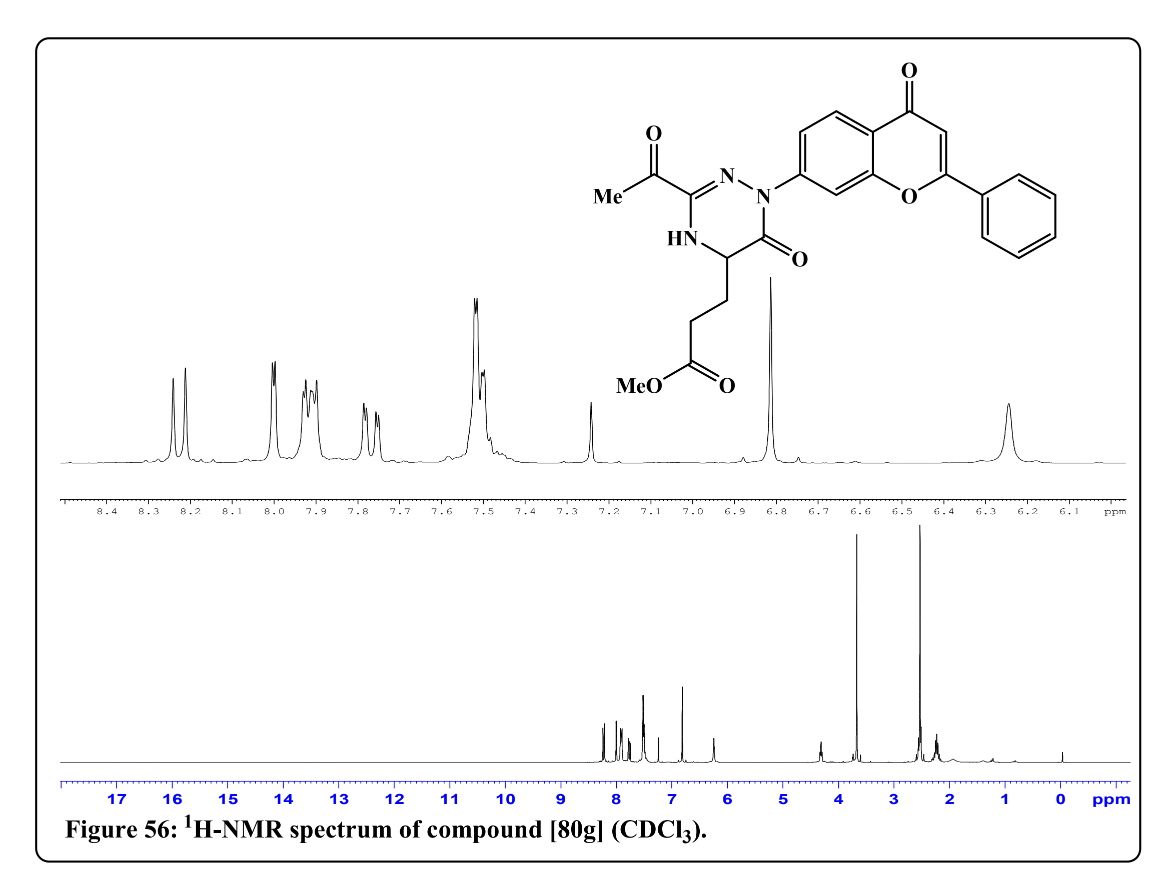


Figure 56: ^{1}H-NMR spectrum of compound [80g] ($CDCl_3$).

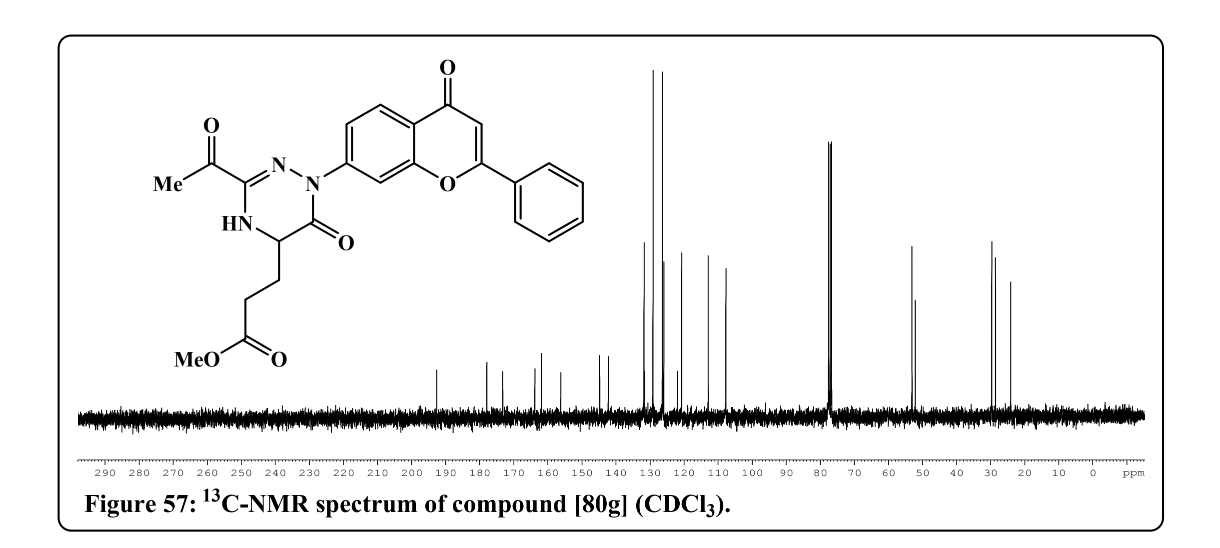


Figure 57: ^{13}C-NMR spectrum of compound [80g] ($CDCl_3$).

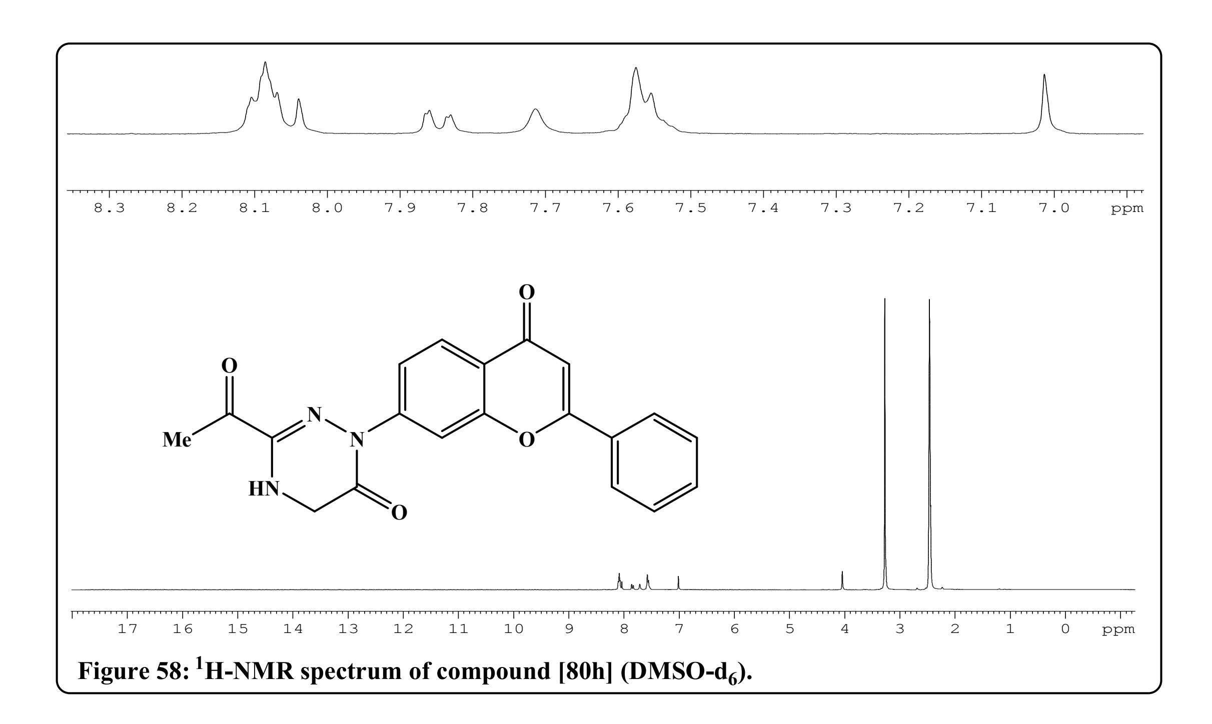


Figure 58: ^{1}H-NMR spectrum of compound [80h] (DMSO-d_6).

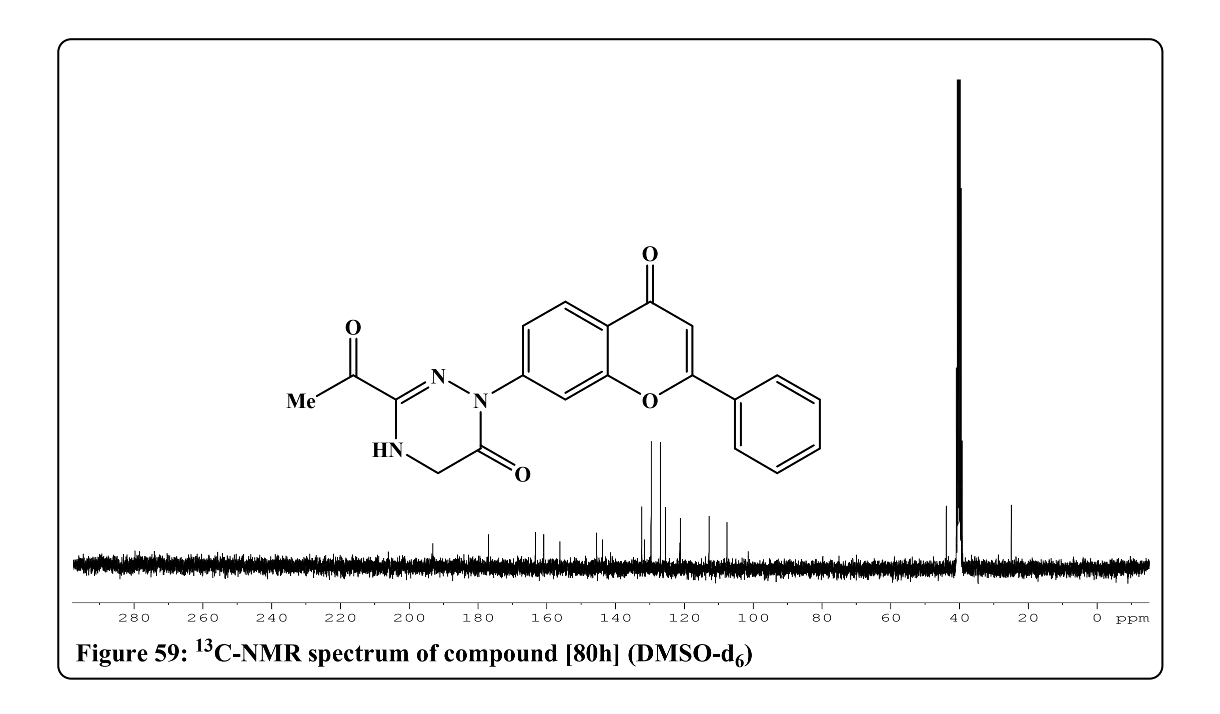


Figure 59: ^{13}C-NMR spectrum of compound [80h] (DMSO-d$_6$)

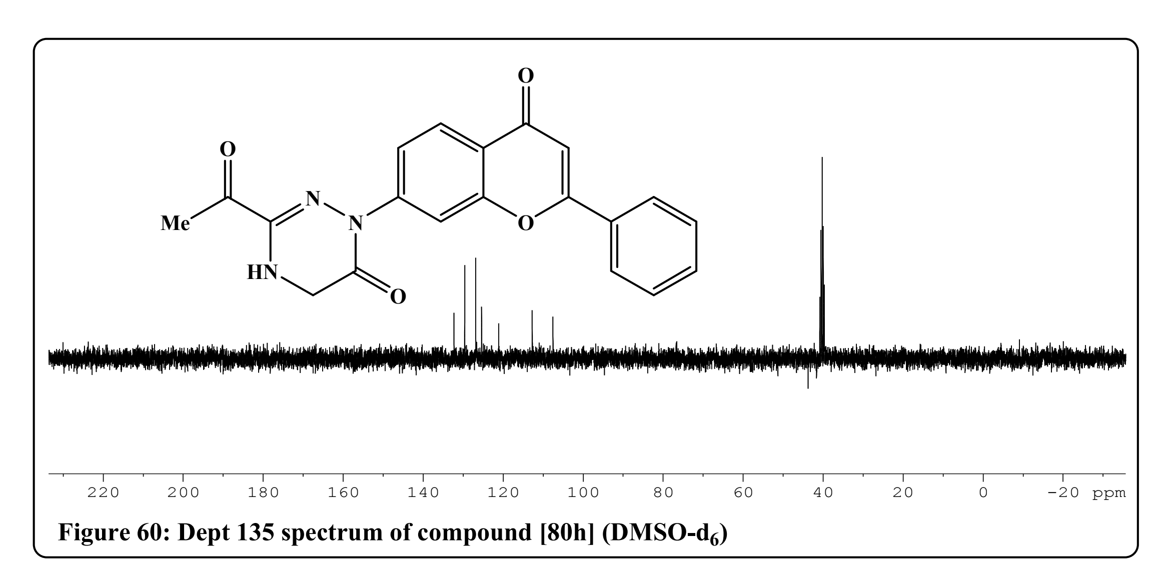


Figure 60: Dept 135 spectrum of compound [80h] (DMSO-d$_6$)

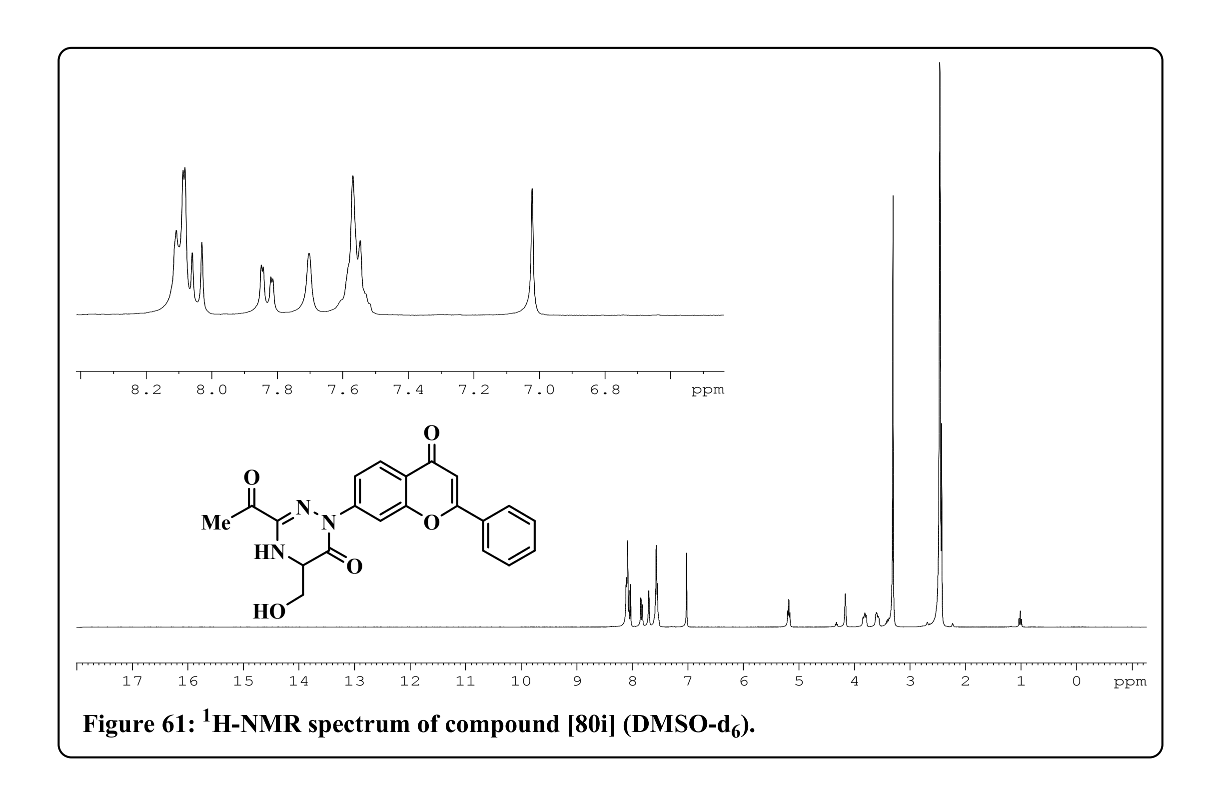


Figure 61: ^{1}H-NMR spectrum of compound [80i] (DMSO-d_6).

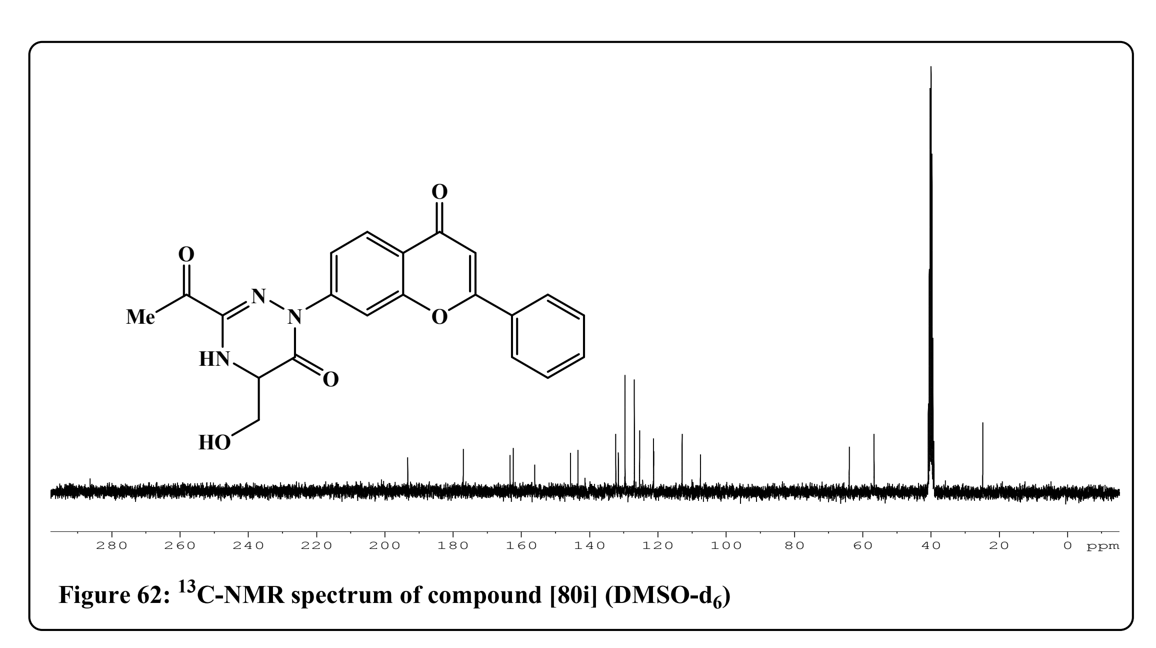


Figure 62: ^{13}C-NMR spectrum of compound [80i] (DMSO-d_6)

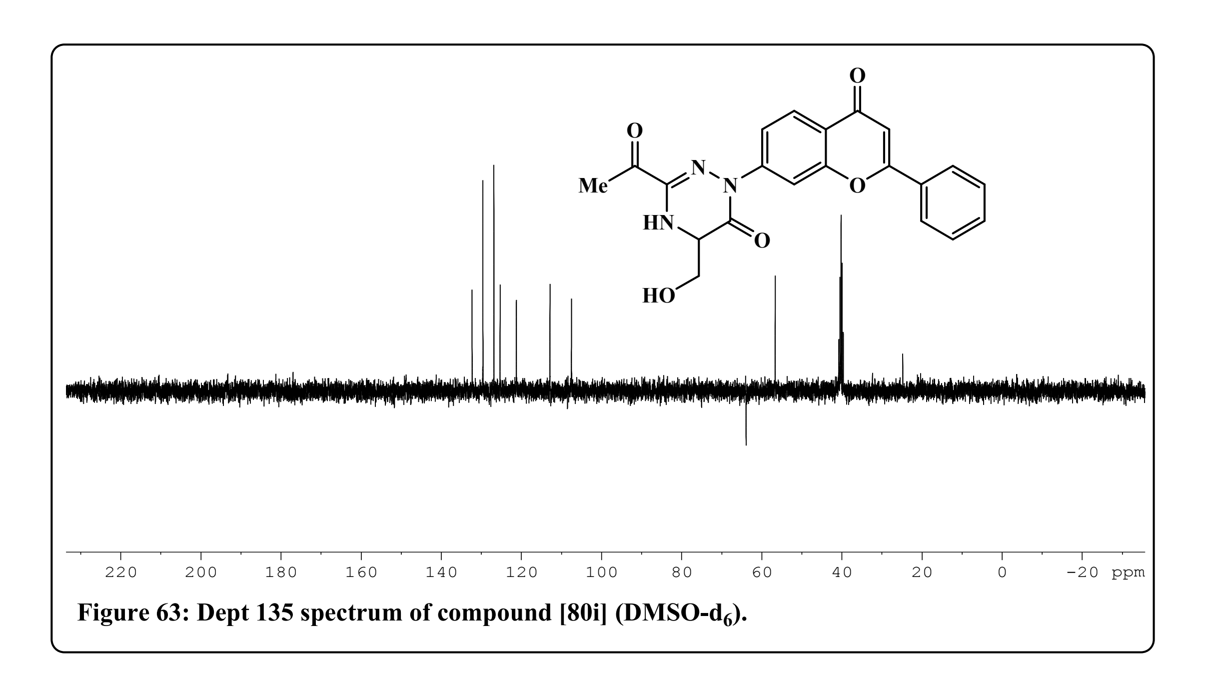


Figure 63: Dept 135 spectrum of compound [80i] (DMSO-d$_6$).

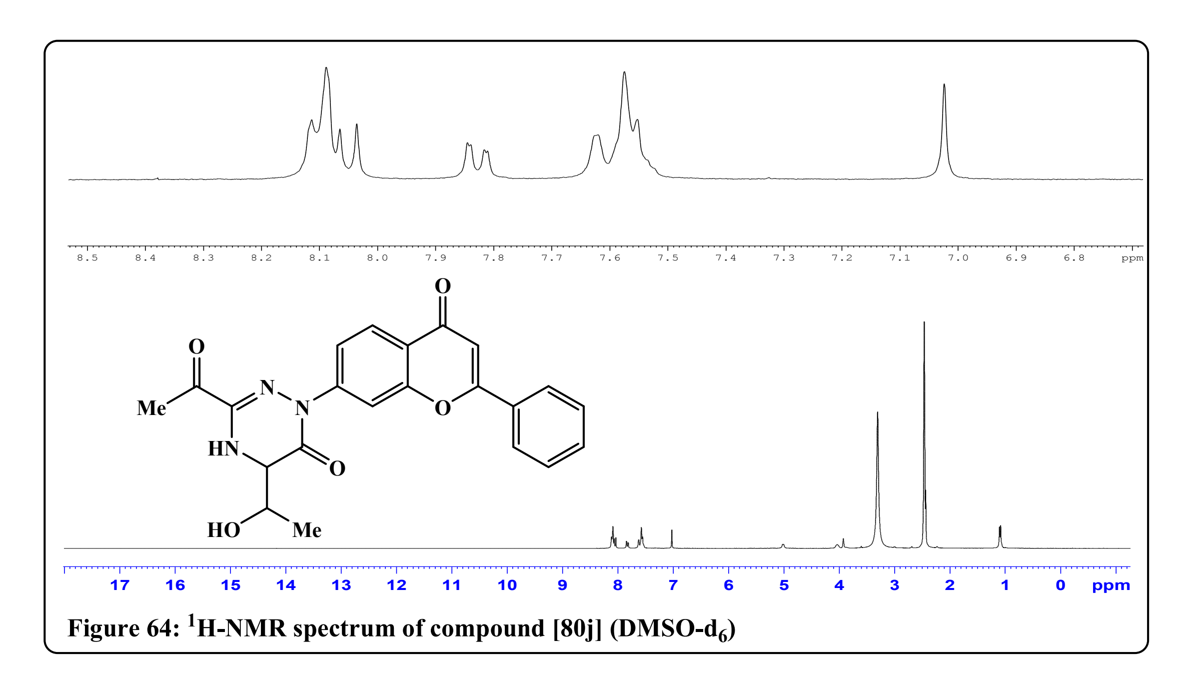


Figure 64: ^{1}H-NMR spectrum of compound [80j] (DMSO-d$_6$)

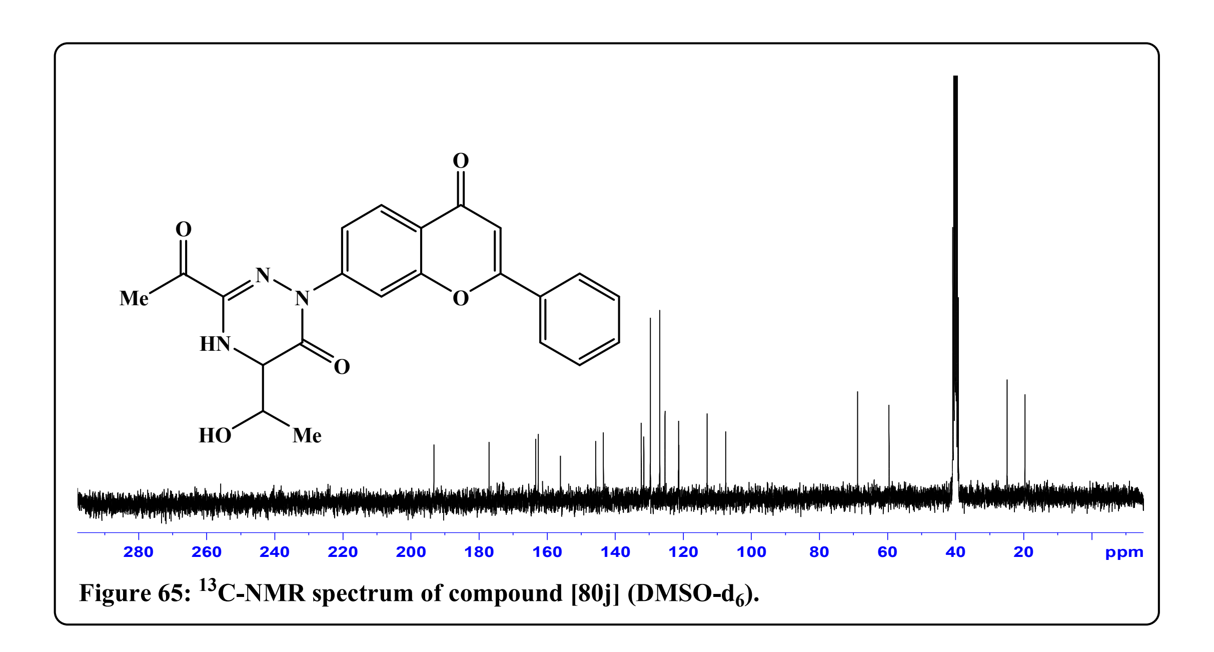


Figure 65: ^{13}C-NMR spectrum of compound [80j] (DMSO-d_6).

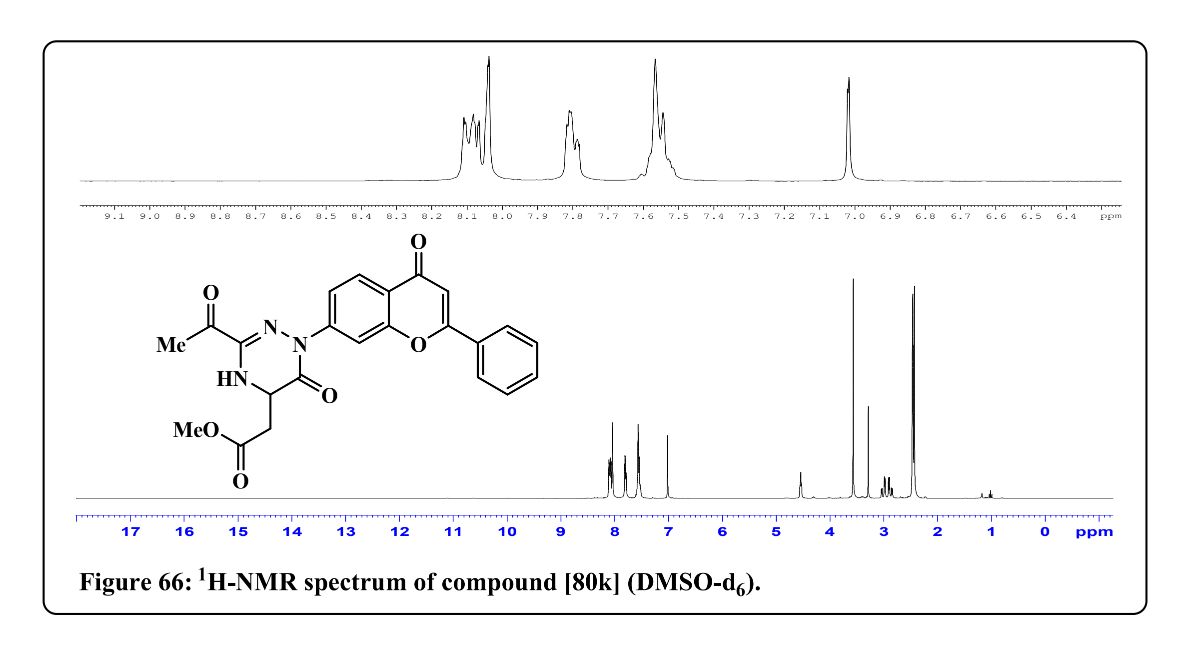


Figure 66: ^{1}H-NMR spectrum of compound [80k] (DMSO-d_6).

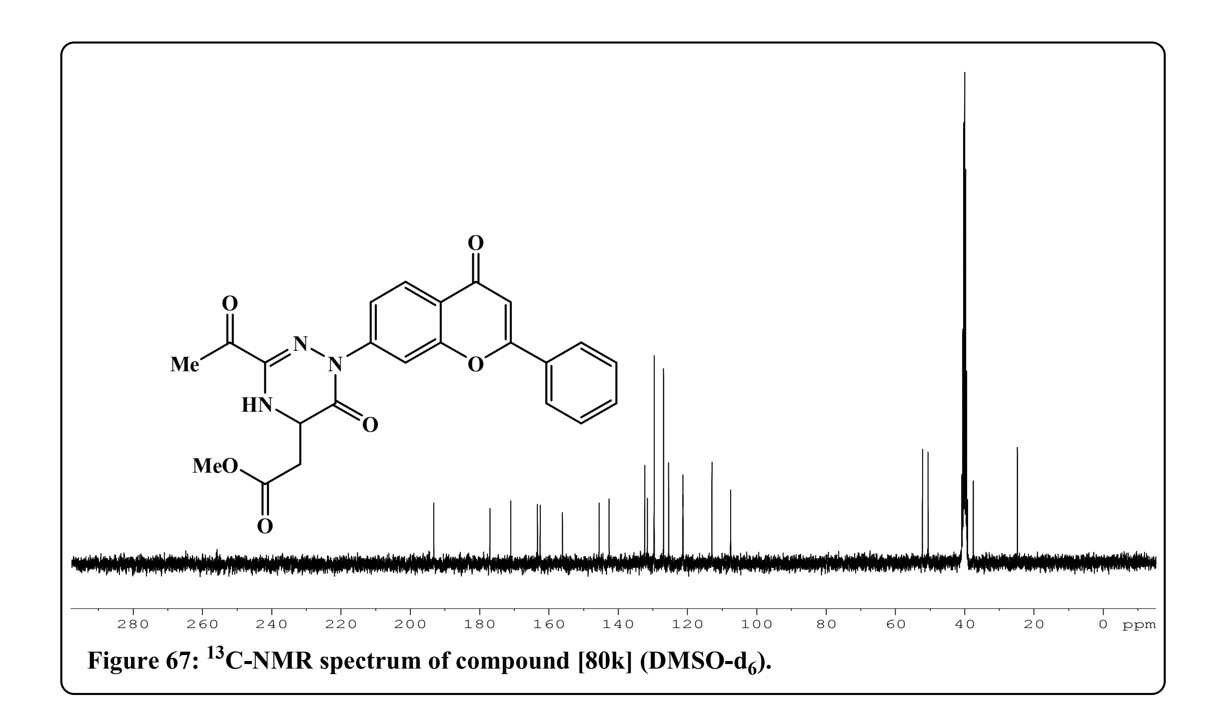


Figure 67: ^{13}C-NMR spectrum of compound [80k] (DMSO-d_6).

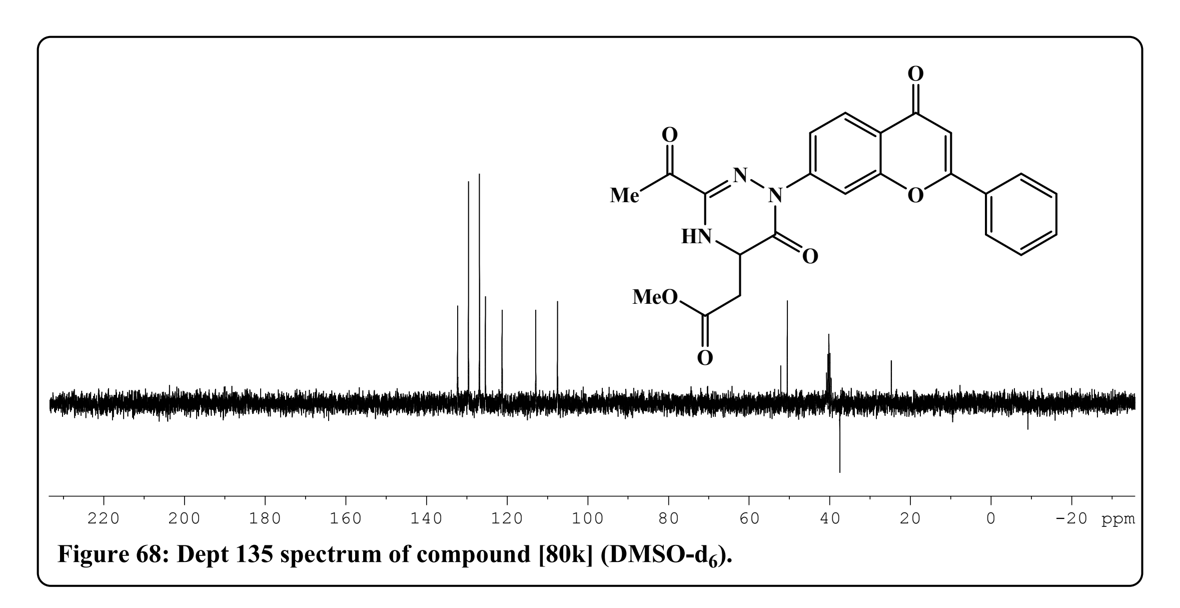


Figure 68: Dept 135 spectrum of compound [80k] (DMSO-d_6).

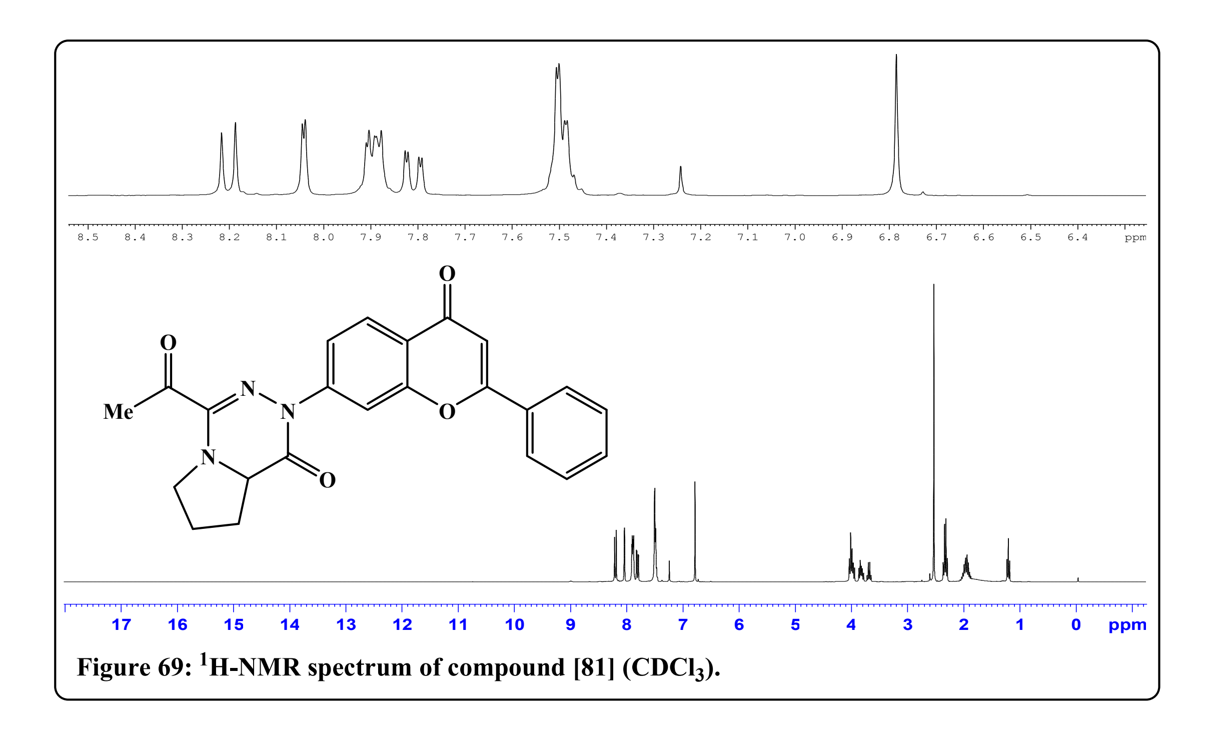


Figure 69: ^{1}H-NMR spectrum of compound [81] ($CDCl_3$).

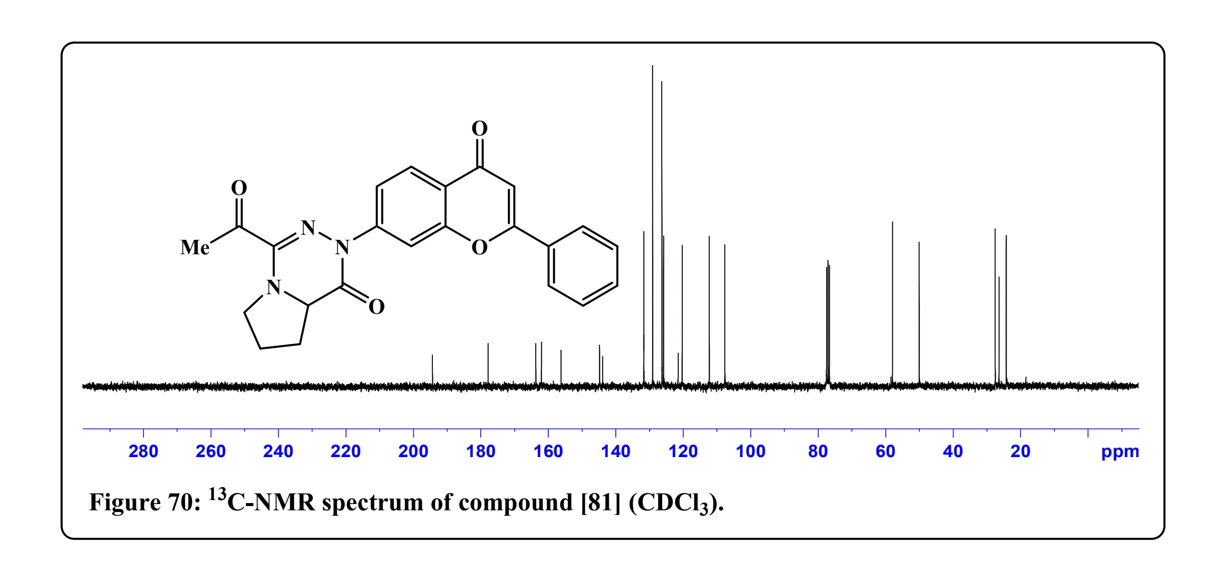


Figure 70: ^{13}C-NMR spectrum of compound [81] ($CDCl_3$).

تحضير ودراسة التأثيرات البيولوجية لبعض مركبات ن1-(فلافون-7-يل)أميدرازون ومثيلاتها ذات العلاقة.

إعداد

مروه نعيم أبو عيشة

المشرف

الأستاذ الدكتور محمد سليمان مبارك

المشرف المشارك

الأستاذ الدكتور مصطفى محمد العبادلة

الملخص

تم تحضير فلافون-7 -يل هيدرازون الكلور (**78**) عن طريق تفاعل جاب -كلنجيمان، بدءا من -7 أمينو -فلافون .و تم مفاعلة الهيدرازون الجديد مع مجموعه مختارة من الأمينات الثنائية في وسط قاعدي ليتحول الى أميدرازونات الفلافون -7 -يل المناظرة (**79**).

و كذلك تم تحضير مركبات(1, 2-4) تريازين-6- ونات الفلافون-7-يل (**80**) و مركب بيرولو(1, 2-د)(1 ,2-4) تريازين-1-ون (**81**) من خلال مفاعلة فلافون-7 -يل هيدرازون (**78**) مع مجموعة من إسترات الاحماض الأمينية بالظروف نفسها.

وقد تم تشخيص هذه المركبات الجديدة باستخدام بعض التقنيات الطيفية كطيف الرنين النووي المغناطيسي للهيدروجين والكربون 13 وتجارب الرنين ثنائية الأبعاد و مطياف الكتلة .و لقد أجري فحص أولي على فعالية المركبات ضد خلايا سرطان الدم و الثدي و أعطت بعض المركبات نتائج جيده إلى ممتازه.